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NUTRIENT-INDUCED CHANGES IN THE SPECIES COMBINATION OF MEADOW ASSOCIATIONS IN AN IRRIGATED SOLONCHAK-SOLONETZ SOIL IN THE DANUBE VALLEY

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(Received September 10, 1966)

The more and more intensive drainage and drying up of the saline inland-watered areas of the Danube valley has caused, since the nineteen-twenties, a progressive transformation of vegetation (HERKE, 1962). The xero-halophilous vegetation, spreading over a larger and larger area, reflects the mosaic-like soil changes depending upon the inland-water conditions. The species combinations of associations taking place here demonstrate, depending upon the cropland conditions, considerable differences concerning both their level configurations and species numbers. The flora of the less saline soils with arranged water-system is the most developed, protruding island-like from the level of watered flats. On the other hand, the species combinations of associations observed in the solonchak and solonchak-solonetz soils in the deeper-sited zone are the poorest, containing at most not more than three-four species as a consequence of the salt accumulation on or near the surface.

The question was raised how to produce multilevelled grasslang-cenoses which would be more developed than the saline meadow, resp. pasture associations *Lepidio-Puccinellietum*, resp. *Lepidio-Camphorosmetum* which dominate large areas at present. The experimental phytocenology may approach the solution of problem in two ways:

(1) By making the impacts considered to be satisfying (soil-amelioration, fertilizer-administration, irrigation, as well an aversowing parallel with them), starting from the existing saline meadow cenoses.

(2) By forming new phytocenoses in the place of existing cenoses, apart from employing the above mentioned treatments.

It is obvious that the most adequate answer to the raised question can be given if started in both ways together. In this paper we want to give an answer only concerning the way outlined in point 2.

The changes in cenoses of meadow associations as a result of fermentizer doses, resp. their measuring on an experimental basis, have been treated of by several authors, so far (KRISTE and WALTER (1955); KLAPP (1927, 1962). The ef-

fect of mineral fertilizers on the *Papilionaceae* and *Gramineae* components was examined by CORDUKES and co-workers (1955) and ECKSTEIN (1934). The influence of nitrogen on the same species groups by GRANT and BROWN (1961); in case of meadow associations by KREIL and co-workers (1961). BALÁZS (1961) has dealt with the importance of lawn fertilization. The geobotanical connections of the changes of natural lawn associations induced by fertilizer administration are treated of by the papers of SIEBOLD (1958) and RUMBURG and COOPER (1961). The effects of fertilizer doses at meadow associations, in case of different cropland factors, are described by SALVADORI (1954) and SCHECHTNER (1961). The general connections are summed up by SIEBOLD (1958).

First I published some data concerning the synecological changes of meadows of saline soil in the Danube valley as a result of different fertilizers (1958); And about similar connections of hayfields in solonetz soils the experimental results from H o r t o b á g y give a picture (1960, 1962).

Some results of lawn cenosis valuation by recent methods were obtained at the Danube sector of Fülöpszállás where we tried to valuate the quantity of production, as well, apart from the changes of dominance relations at any species (HARMATI—BODROGKÖZY, 1965; BODROGKÖZY—HARMATI, 1966).

Methods

For clearing up the questions raised in connection with the change of meadow association of solonchak-solonetz stilts, an experiment series was arranged in the Experimental Settlement for sodic soil amelioration in S z u n y o g p u s z t a (Kiskunlacháza) in the framework of a scientific cooperation program of the Institute for Botany of the Attila József University and the Agricultural Experimental Institute of the South-Hungarian Plain, in the Autumn of 1963.

The treatments of the four series experiment, arranged according to the split-plot system, comprised both the possibilities of the amelioration of alkali soils and tohose of the fertilizer administration. Their combination proved to be:

(1)	a_0 b_0	(5)	a_1 b_0	(9)	a_2 b_0
(2)	a_0 b_1	(6)	a_1 b_1	(10)	a_2 b_1
(3)	a_0 b_2	(7)	a_1 b_2	(11)	a_2 b_2
(4)	a_0 b_3	(8)	a_1 b_3	(12)	a_2 b_3

Soil-amelioration (Autumn 1963)

a_0 = unameliorated
a_1 = 27 q $CaSO_4 \cdot 2H_2O$/ha
a_2 = 67 q $CaSO_4 \cdot 2H_2O$/ha

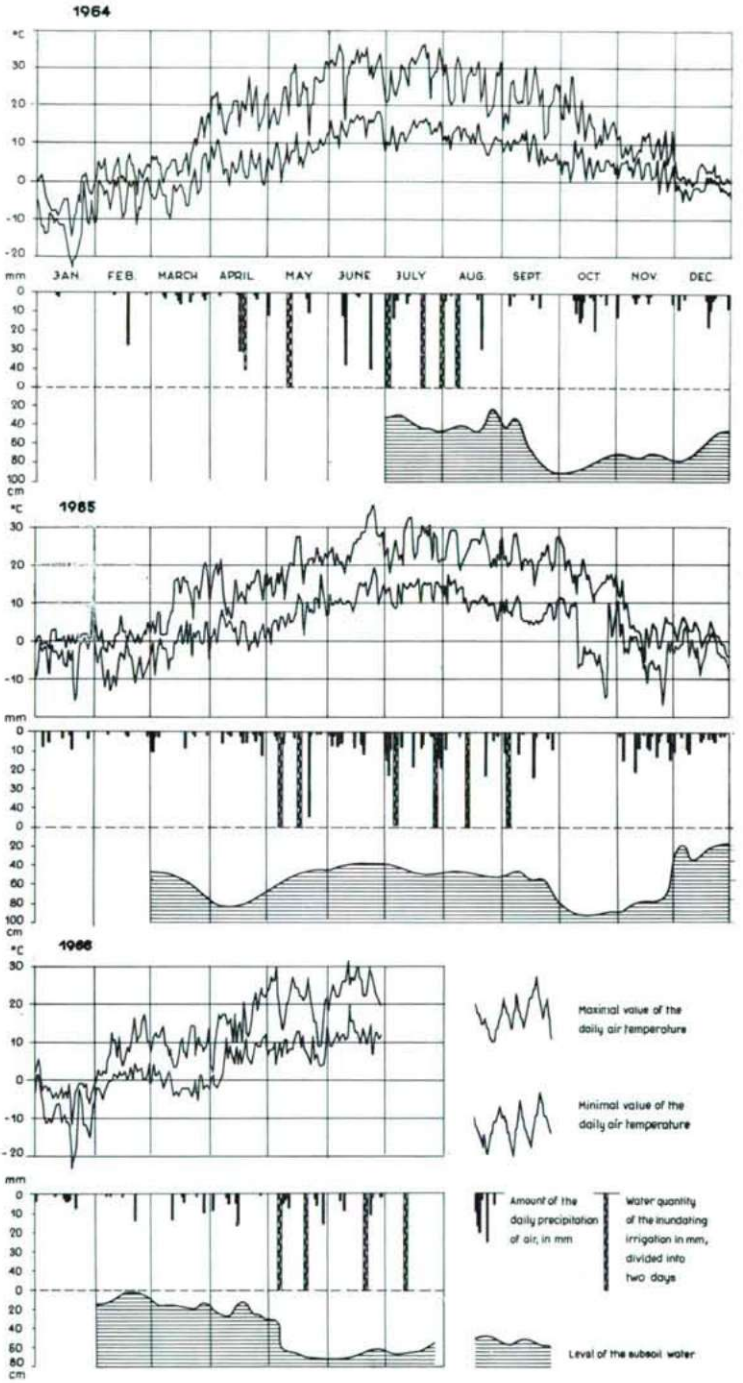
Fertilization (repeated yearly)

b_0 = unfertilized
b_1 = 90 kg P_2O_5/ha
b_2 = 74 kg $NH_4 \cdot NO_3$/ha
b_3 = 90 kg P_2O_3+74 kg $NH_4 \cdot NO_3$/ha

At setting the experiment, in August 1963, after the original saline meadow of *Lepidio-Puccinellietum asteretosum* type had been broken by a distiller, sulphate of lime and fertilizer doses got into the soil, with a seed mixture composed apropriately to the subject.

The alkali plains in the Danube valley dry up more and more in a natural way or by reclaiming and the salt-concentration of soil increases. For eliminating this harmful effect and for ensuring the water required by the hygrophilous species, we have made possible the intermittent irrigation on the experimental plots by inundation and by surface irrigation. In dry, rainless periods we administered the inundation water 3—4 times in every vegetation cycle,

Fig. 1. Change of temperature of the amount of precipitation, of irrigation water, and of the level of subsoil water during the period of examination.


1964
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cm
Maximal value of the daily air temperature
Minimal value of the daily air temperature
Amount of the daily precipitation of air, in mm
Water quantity of the inundating irrigation in mm, divided into two days
Level of the subsoil water

according to 100 mm atmospheric precipitation. The excess of water not absorbed by the soil (40—60 per cent or so) was drained from the area.

The use of components in a greater number than in routine procedures was justified by the supposed selective influence of the solonchak-solonetz soil where, for the time being, the degree of selection is more or less unknown.

For determining the special amount of seed used for sowing, we kept in mind the probable degree of the concurrence between the species. Some date concerning their influence are published by ARENS (1962). The components are species requiring different croplands and forming different grass levels. Their distribution is as follows:

	Lower	Middle	Upper	Seed	
	grass level			kg/ha	per cent
Molinietalia and *Arrhenatheretea* species:					
Arrhenatherum elatius			0	5,91	17,0
Phleum pratense			0	0,06	2,8
Trifolium hybridum		0		0,70	2,0
Lotus corniculatus	0			3,39	9,7
Agrostion species:					
Festuca pratensis			0	2,35	6,7
Alopecurus pratensis			0	1,22	3,5
Beckmannion species:					
Beckmannia eruciformis			0	2,61	7,5
Bromion and *Festuco-Brometea* species:					
Poa angustifolia		0		2,26	6,5
Bromus inermis			0	8,34	24,0
Species indifferent to cultivation and association:					
Lolium perenne		0		1,51	4,3
Medicago sativa		0		3,75	10,8
Festuca rubra	0			1,82	5,2

The daily precipitation distribution of the firat three years of the experimental priod, as well the dates and amounts of irrigations and the change of maximum-minimum values of the air temperature as measured on the grass level ar demonstrated by Fig. 1 together with the level curve of subsoil water.

The qualitative and quantitative changes of the association conditions of experimental plots were fixed two times in each vegetation period, in the Spring and Autumn aspects, immediately before mowing time, in the form of phytocenologic surveys, giving the values of D-scale in percentage.

The mean height of species was ascertained, as well, and by the help of it also the calculation of the supersoil production amount could be carried out for every species. We can find general productionecologic examinations at GORHAM and PEARSAL (1956); meadow production examination results by phytocenologic surveys in BALÁZS's papers (1944., 1949), as well in the

works of IVINS (1959) and SEARS (1962). And data concerning the plant ecologic examinations of their productivity and quality are published by UBRIZSY (1943).

Our method employed in this area — which is simpler and can be carried out faster — was demonstrated at the Symposium of the International Society for Plant Geography and Ecology in 1966 and applied at a number of similar examinations of ours, giving good connections between the total overground production amount of plots and the weight of dry-hay.

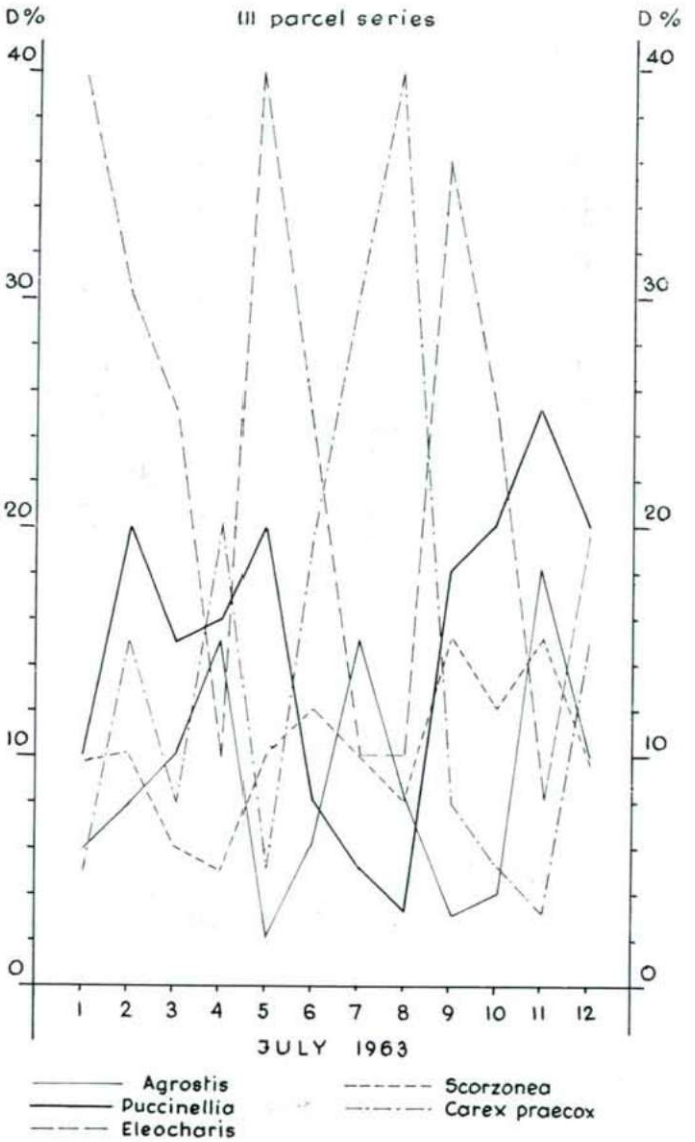


Fig. 2. Change of dominance percentage of major species of the original plant association in parcel series III (parcels 1—12) of the experimental area.

Original vegetation of the experimental plots

For making clear and evaluating the connections of the synecologic conditions of meadow associations, their detailed phytocenologic croplandecologic analyses and examinations are indispensable. We obtain some data for their comprehensive knowledge from KLAPP (1951); HUNDT (1958); MARGIT KOVÁCS (1958), VICHEREK (1962), and — concerning the area between Danube and Tisza — from BODROGKÖZY (1960).

The results of the synecologic analyses of the saline meadows in the Danube valley are going to be published in the next future, the most extensive association of their beloise-eroded saline plains being *Lepidio-Puccinellietum.* They are cultivated here and there with inundating irrigation for growing their yield. Their solonchak-solonetz soils got thereby more and more disalkalized, their Na_2CO_3 content decreased. The typical form of the original *Puccinellia* meadow grew richer with mildly halophytic hygrophilous species. Thus its species combination was entered by *Agrostis alba, Eleocharis uniglumis, Aster tripolium ssp. pannonicus, Rorippa silvestris ssp. kerneri.* This periodic inundation, however, could not prevent thoroughly the single xerophilous halophytae from occuring, and the *Hordeum hystrix Scorvonera cana* was even multiplied considerably. An interesting phenomenon is the sporadic multiplication of *Carex praecox* having an extremely high ecologic adaptability; its dominance values can reach even 40 per cent.

Our experimental plots happenned to lie similarely in such saline meadow sections inundated for a long time. Their plant substance was rather uniform in a larger area and, at the beginning of the experiments, the original vegetation could be considered as a withering version of *Lepidio-Puccinellietum limosae asteretosum,* forming, at any rate, very extensive stocks in the Danube valley and being more and more a burdon on the agricultural practice.

The changes of dominance and quantitative output of the original vegetation in the single plots are demonstrated on the original species combinations of repetition III of the experimental series, divided according to plots (Fig. 2).

Results of the first vegetation year:

The first cenologic surveying of our experimental plots could take place in the Spring aspect after plantation, i. e., in June 1964.

From the four experimental series, a survey and measurements of mean height of repetition I and III were carried out according to plots and species.

(1) As the exact establishment of the springing and strengthened species and even the measurement of their dominance values, could be performed, we got the surprising results, unexpected for us, that 75 per cent of the planted species occurred in the species combinations of plots in spite of the saline soil conditions.

(2) In the frist vegetation year the following characteristics can be established:

(a) There couldn't be discovered, as yet, any considerable difference between the plots ameliorated by a fertilizer and the control plots which were not fertilized. Therefore, we treat in the following only of the evaluation of changes that occurred as a consequence of the fertilizer doses (Tab. I).

As a result of the breaking up, some of the species of the original meadow association got back among the sowed species, a leading role, however, could be obtained first of all by the ephemerous halophytic *Puccinellietalia* elements like *Hordeum hystrix, Matricaria chamomilla var. salina, Cerastium dubium,* and also the degree of return of *Puccinellia limosa* is considerable; the output of its production is 6—12 at a 5—10 per cent cover.

(b) From the sowed glycophytae the association-indifferent *Lolium perenne* showed up the highest overlay value; its quantitative output is nearly always over 10. Under saline conditions a special interest may be aroused by the occurence of *Arrhenatherum elatius* whose natural meadow associations were examined by SCHNEIDER (1954), and its ecologic borders were treated of by APERDANNIER (1959). From the *Papilionaceae* species the values of the *Molinietalia* elements, of *Trifolium hybridum* and *Lotus corniculatus* were surpassed by the values of *Medicago sativa* at the beginning.

(3) (a) On plots fertilized with P_2O_5 (90 kg/ha), from the grass species first of all *Lolium perenne* was influenced considerably, showing a 100 per cent increase compared with unfertilized plots considering both the dominance and the quantitative output. If influenced by Phosp-

TABLE I

Unfertilized parcels June 3rd 1964.

Fertilizer and its amount:	∅				$CaSO_4 \cdot 2H_2O$							
					27 q/ha				67 q/ha			
Number of parcel series:	I.		III.		I.		III.		I.		III.	
Number of all species:	17		17		14		15		15		11	
Dominance and production amount:	%	P	%	P	%	P	%	P	%	P	%	P
Puccinellion and Puccinellietalia species:												
H Puccinellia limosa	6	6,9	8	9,6	5	5,8	8	11,6	8	9,8	10	12,3
Th Hordeum hystrix	15	5,2	15	7,2	10	3,4	20	7,6	10	3,0	10	3,4
Th Cerastium dubium	25	8,7	5	1,4	15	4,9	5	1,5	10	2,8	2	0,6
Th Matricaria chamomilla var. salina	18	2,7	12	2,0	10	1,5	18	3,4	17	3,6	10	2,0
Festucion pseudovinae species:												
H Scorzonera cana	0,5	0,1	0,5	0,2	·	·	·	·	0,5	0,1	·	·
Agrostion species:												
H Festuca pratensis	1	1,7	1	1,6	2	3,5	1	1,8	3	4,9	3	5,2
H Alopecurus pratensis	·	·	1	1,7	0,5	0,8	1	1,7	0,5	0,8	·	·
Th Ranunculus sardous	0,2	0,5	·	·	·	·	0,5	1,4	·	·	·	·
H Agrostis alba	1	1,1	2	2,5	·	·	·	·	·	·	·	·
Molinietalia and Arrhenatheretea species.												
H Trifolium hybridum	4	2,3	2	1,1	2	1,1	1	0,6	1	0,6	1	0,5
H Lotus corniculatus	2	1	1,5	0,7	0,5	0,2	0,5	0,2	0,5	0,2	1	0,5
H Arrhenatherum elatius	3	4,1	10	1,4	2	2,6	5	6,2	6	8,0	6	8,2
Bromion-, Festuco-Brometea species:												
H Bromus inermis	0,5	8,0	1	1,6	0,5	2,0	0,5	1,8	0,5	0,7	·	·
H Poa angustifolia	0,5	0,5	0,5	0,6	1	1,2	0,5	0,6	1	1,2	1	1,2
Th Bromus mollis	1	1,2	1	1,3	·	·	1	1,2	·	·	·	·
Association-indifferent species:												
H Medicago sativa	4	1,7	3	1,4	5	1,0	1	0,4	2	0,9	2	1,0
Th Lolium perenne	10	10,3	15	17,2	10	9,2	15	15,0	10	10,7	15	14,4
Th Polygonum aviculare	0,5	·	1	0,1	1	0,1	·	·	1,5	0,2	·	·

phorus, also the *Arrhenatherum* continued increasing its advantage. At the same time, the cover of the thereophytic *Puccinelietalia* elements, first of all that of the species *Matricaria chamomilla v. salina* and *Cerastium dubium* decreased in a considerable degree — while their number did increase with the *Atriplex litoralis* and *Myosurus minimus* occurring thread by thread. *Hordeum hystrix* is, however, an exception.

From the *Papilionaceae* we have expected more considerable advance, however, only *Trifolium hybridum* demonstrated some increase concerning the D percentage. Concerning the effects of Phosphorus-doses on the *Trifolium* species we may, anyway, find some recent data at GERVAIS (1960), and- in grass papilionaceae combination- at HUNG and WAGNER (1963).

(b) The upper grass level of the phosphorus-treated plots of the produced hayfield was et most still open in the Spring aspect of the first year. *Arrhenatherum* prevailed. The highest closing value was reached by the middle grass level, its dominating species being *Lolium perenne, Puccinellia limosa, Trifolium hybridum.* The lower grass level got under the dominance of *Hordeum hystrix* (Tables II, III).

(4) The influence of NH_4NO_3 (174 kg/ha) is advantageus first of all from the point of view of grass species; especially in case of *Festuca pratensis* whose development was investigated by SEREBYAKOVA (1962). While hardly any P-effect could be demonstrated on it in the first vegetation year, after influenced by N, the percentage of D and the value of quantitative output grew double, on the average.

(b) The species of the produced meadow association demonstrated an increased closing in al the three grass levels as a result of nitrogen. The question whether the prevalence or withdrawal of the single species is influenced more by the effect of fertilizer doses or by the conditions of illumination influenced by the closing (SPEIDEL 1966) or rather by the increasing struggle for life of species can hardly be answered exactly, and we must not forget the selective effect of the special physical and chemical influence of the soil, either.

(c) For characterizing the single grass levels, we must ascertain that already in the first vegetation year sharp differences could be seen concerning both the dominance and quantitative output, to the advantage of nitrogen treatment, opposite to both the control and the P-treated plots. The 40—50 per cent closing of the upper grass level is a result of the summing up of dominance values of *Arrhenatherum elatius* (20—30 p. c.), *Festuca pratensis* (5—12 p. c.), and *Bromus inermis* (3—7. p. c.). We expected the last mentioned species in the formation of the upper level to get the leadership as it was for years the dominant species of the level in the solonetz soil H o r t o b á g y as a result of fertilizer doses (BODROGKÖZY 1962), and 24 per cent (8,34 kg/ha) of the grain sowed in the plots was obtained from *Bromus inermis.*

The dominant species of the middle grass level was *Lolium perenne,* with a further considerable expansion although yielding only, 4,6 per cent of the sown grass seed. Beside it is *Puccinellia limosa,* without any major change. The *Papilionaceae* of that level sufferred an extremely great regression; they survived hardly, only thread by thread, rather spindled for want of water.

The species of the lower grass level were obtained, also henceforward, from the ephemerous species of *Puccinellietalia* elements; first of all *Hordeum hystrix* and *Cerastium dubium* are rather considerable, at which the nitrogen effect, independently of the increased overshadowing, caused a considerable expansion. The values of *Matricaria* have not changed considerably (Fig. 3).

TABLE II

P_2O_5 (90 kg/ha) treated parcels June 3rd 1964.

Fertilizer and its amount:	∅				$CaSO_4 \cdot 2H_2O$							
					27 q/ha				67 q/ha			
Number of parcel series:	I.		III.		I.		III.		I.		III.	
Number of all species:	15		16		13		14		14		13	
Dominance and production amount:	%	P	%	P	%	P	%	P	%	P	%	P
Puccinellion- and Puccinellietalia species:												
H Puccinellia limosa	8	9,1	5	5,8	10	11,9	8	10,5	5	5,8	5	0,6
Th Hordeum hystrix	10	6,9	8	4,6	12	7,4	10	5,8	15	8,6	5	3,0
Th Cerastium dubium												
Th Matricaria chamomilla var. salina	2	0,7	1	0,3	5	1,4	1	0,3	3	1,1		
Th Atriplex litoralis		0,5							1	0,3		
Th Myosurus minimus	0,5	0,3										
Festucion pseudovinae species:												
H Scorzonera cana					0,5	0,2						
Beckmannion species:			1									
H Beckmannia eruciformis			0,5	1,2	1	1,2	1	1,2			1	1,2
H Rorippa silvestris ssp. kerneri	0,5	1,4		1,4								
Agrostion species:			4									
H Festuca pratensis	4	3,8		11,2	3	3,5	3	3,7	5	5,8	2	1,9
H Alopecurus pratensis			2									
H Agrosis alba	1	0,8		1,7			2	1,5	1	0,2	1	0,8
Molinietalia and Arrhenatheretea species:			5									
H Trifolium hybridum	3	0,9	3	1,7	2	0,7	2	0,6	3	0,9	3	1,0
H Lotus corniculatus	1	0,3	8	0,3	1	0,3	1	0,3	1	0,3	1	0,3
H Arrhenatherum elatius	10	15,3	2	11,5	16	23,6	8	11,1	10	15,3	8	11,0
H Phleum pratense	1,5	12,9		1,5			2	1,6	1	0,7	2	1,6
Bromion and Festuco-Brometea species:												
H Bromus inermis	1	1,3	4	4,6	1	1,2	1	1,3	1	1,1	2	1,3
Th Bromus mollis	0,5	0,4	1	1,0	1	0,6					0,5	0,3
H Poa angustifolia			1	1,3			1	1,2				
Association-indifferent species:												
H Medicago sativa	3	1,4	3	1,4	5	1,1	2	0,1	2	0,1	1	0,4
H Lolium perenne	30	34,5	15	17,3	25	27,8	20	23,8	30	32,2	20	24,9
Th Polygonum aviculare									0,5	—		

TABLE III $NH_4 \cdot NO_3$ (174 kg N/ha) treated parcels June 3rd 1964.

Fertilizer and its amount	∅				$CaSO_4 \cdot 2H_2O$							
					27 q/ha				67 q/ha			
Number of parcel series:	I.		III.		I.		III.		I.		III.	
Number of all species:	14		12		12		16		12		17	
Dominance and production amount:	%	P	%	P	%	P	%	P	%	P	%	P
Puccinellion Puccinellietalia species:												
H Puccinellia limosa	10	13,8	8	10,7	8	1,2	8	10,7	8	11,0	5	6,7
Th Hordeum hystrix	5	3,8	12	9,7	13	10,0	12	9,9	15	12,0	8	6,1
Th Cerastium dubium	5	1,9	10	4,2	12	5,0	10	4,4	3	1,2	5	1,9
Th Matricaria chamomilla	3	0,6	5	0,1	5	0,1	1	0,3			2	0,6
Th Atriplex litoralis	·	·	2	0,5	·	·	2	0,7	·	·	1	0,3
Festucion pseudovinae species:												
H Scorzonera cana	·	·	·	·	0,5	0,1	·	·	0,5	0,1	·	·
Beckmannion species:												
H Beckmannia eruciformis	3	4,0	·	·	2	2,7	1	1,3	1	1,4	1	1,4
Agrostion species:												
H Festuca pratensis	10	13,4	8	10,0	6	8,0	9	12,4	8	11,2	5	6,7
H Alopecurus pratensis	3	4,3	2	2,9	2	2,9	3	4,1	3	4,0	4	5,5
H Agrostis alba	·	·	·	·	·	·	1	1,1	·	·	1	1,1
Molinietalia and Arrhenatheretea species:												
H Arrhenatherum elatius	20	3,4	24	42,3	25	43,1	20	3,2	30	52,9	25	43,1
H Phleum pratense	2	2,5	·	·	·	·	1	1,2	3	3,4	2	2,5
H Trifolium hybridum	·	·	0,5	0,1	·	·	·	·	·	·	·	·
H Lotus corniculatus	·	·	·	·	·	·	0,5	0,1	·	·	0,5	0,1
Bromion and Festuco-Brometea species:												
H Bromus inermis	6	7,5	3	3,7	5	5,9	6	7,5	6	7,7	5	6,2
H Poa angustifolia	1	1,2	·	·	1	1,2	·	·	·	·	·	·
Th Bromus mollis	1	1,2	·	·	·	·	·	·	1	1,2	1	1,2
Association indifferent species:												
H Lolium perenne	30	37,4	25	3,4	20	27,6	22	27,5	20	28,8	30	39,1
H Medicago sativa	·	·	·	·	·	·	0,5	0,2	·	·	0,5	0,2
Th Polygonum aviculare	1	0,1	2	0,2	·	·	1	0,1	·	·	2	0,2

(5) (a) In the first year of vegetation, the double-treated (P + N) plots showed up meadow-association conditions similar to those treated with nitrogen. At some species components, however, some differences could be observed that required consideration. Thus, inside the double treatment, and concerning the demand on phosphorus, it seems so already in the first phase of experiments that the original P-content of soil is enough for the undisturbed development of *Festuca pratensis*, and a further P-amount is of negative effect in consideration of both the covering percentage and the quantitative output (Fig. 3). On the other hand, in case of *Papilionaceae* the N of the double treatment exerts a checking effect.

(b) The influence of P+N—treatment on the grass levels of the produced meadow association manifested itself first of all in the more and more increased closing: the upper level may reach 50—60 and even 70 per cent on the average. From its grass species *Arrhenatherum* keeps leading henceforward, too; the other species don't differ considerably from the solo—N—treated plots.

From the species of the middle level, the *Papilionaceae* have got some advantage compared with the former ones, and also the covering percentage of *Lolium perenne* increased.

Hordeum hystrix shows a further expansion, as influenced by the double fertilizer doses in its lower grass level, altough the degree of overshadowing has continued increasing. It can reach even a 15—20 per cent covering in some plots.

(6) (a) In the Autumn aspect, the differences between the treatments may somewhat grow indistinct, and the differences in Tables concerning the species combinations depend mainly upon the N—treatment. From the levels, the closing of the upper grass level has decreased to be 15—20 per cent; at the same time, *Lolium perenne*, dominating in the middle level, is of a 50—55 per cent cover.

(b) The P—effect is the most intensive from *Papilionaceae* at the *Trifolium hybridum* that springs the fastest after the June mowing, and may reach a 5—8 per cent cover at the end of September, as a result of the propitious light conditions. At the same time, it showed a 3—6 per cent D—value in control plots. *Medicago sativa* has not reached, even with *Lotus corniculatus* together, the 1 per cent coverage averages.

In the untreated and solo—P—treated plots, from the introduced *Puccinellietalia* species, the characteristics of the Autumn aspect, *Aster tripolium ssp. pannonicus*, as well *Artiplex litoralis* have demonstrated a higher coverage value.

Changes taking place in 1965

(1) (a) C l i m a t e : Meadow association in alkali soils and their climates having a close interdependence, it is right if we evaluate the climatic data measured on the grass level, as well. The first part of the vegetation year was favourable for the planted *Agrostion* and *Molinietalia* representatives of our meadow association. From March the climate was cooler, more uniformly rainy than in the former years, decreasing the concentration of soil salts, influenced also by the irrigation. (Fig. 1)

(b) The change of the subsoil level was also in that period of positive effect. The uniform distribution of precipitation, resulted from lacking of a rainfall

TABLE IV

Wet extract

Depth in cm	Ca^{++}	Na^{+}	K^{+}	Mg^{++}	Amount of kations	CO_3^{--}	HCO_3^{-}	Cl^{-}	SO_4^{--}	Amount of anions	Kation + anion mg/100 g
	mg equivalent / mg 100 g					mg equivalent / mg 100 g					
Unameliorated											
0—10	0,29 / 5,81	2,44 / 56,11	0,03 / 1,17	0,47 / 5,71	3,23 / 68,80	0,04 / 1,20	2,56 / 156,21	0,22 / 7,80	0,30 / 14,41	3,12 / 179,62	248,42
10—20	0,10 / 2,00	3,85 / 88,54	0,03 / 1,17	0,08 / 0,97	4,06 / 92,68	0,10 / 3,00	3,19 / 194,65	0,25 / 8,86	0,65 / 31,22	4,19 / 237,73	330,41
20—30	0,10 / 2,00	3,29 / 75,66	0,02 / 0,78	0,16 / 1,95	3,57 / 80,39	0,30 / 9,00	2,49 / 151,93	0,24 / 8,51	0,73 / 35,06	3,76 / 204,50	284,89
30—50	0,07 / 1,40	2,93 / 67,38	0,02 / 0,78	0,01 / 0,12	3,03 / 69,68	0,74 / 22,20	1,76 / 107,39	0,16 / 5,67	0,42 / 20,17	3,08 / 155,43	225,11
50—70	0,06 / 1,20	2,27 / 52,20	0,02 / 0,78	0,09 / 1,09	2,44 / 55,27	0,44 / 13,20	1,54 / 93,97	0,21 / 7,45	0,36 / 17,29	2,55 / 131,91	187,18
70—90	0,04 / 0,80	1,47 / 33,81	0,02 / 0,78	0,09 / 1,09	1,62 / 36,48	0,18 / 5,40	1,11 / 67,73	0,16 / 5,67	0,31 / 14,89	1,76 / 93,69	130,17
Dressed with sulphate of lime (a_1)											
0—10	0,77 / 15,43	1,72 / 39,55	0,02 / 0,78	0,36 / 4,38	2,87 / 60,14	— / —	2,40 / 146,44	0,17 / 6,03	0,34 / 16,33	2,91 / 168,80	228,94
10—20	0,07 / 1,40	3,36 / 77,27	0,04 / 1,56	0,27 / 3,28	3,74 / 83,51	0,30 / 9,00	2,22 / 135,46	0,32 / 11,35	0,88 / 42,27	3,72 / 198,08	281,59
20—30	0,06 / 1,20	3,50 / 80,49	0,03 / 1,17	0,08 / 0,97	3,67 / 83,83	0,26 / 7,80	2,48 / 151,32	0,24 / 8,51	0,70 / 33,62	3,68 / 201,25	285,08
30—50	0,06 / 1,20	2,96 / 68,07	0,03 / 1,17	0,17 / 2,07	3,22 / 72,51	0,34 / 10,20	2,09 / 127,53	0,19 / 6,74	0,62 / 29,78	3,24 / 174,25	246,76
50—70	0,04 / 0,80	2,21 / 50,82	0,04 / 1,56	0,01 / 0,12	2,30 / 53,30	0,50 / 15,00	1,29 / 78,71	0,26 / 9,22	0,44 / 21,13	2,49 / 124,06	177,36
70—90	0,04 / 0,80	1,06 / 24,38	0,02 / 0,78	0,05 / 0,61	1,17 / 26,57	— / —	0,95 / 57,97	0,15 / 5,32	0,21 / 10,09	1,31 / 73,38	99,95
Dressed with sulphate of lime (a_2)											
0—10	1,04 / 20,84	1,28 / 29,44	0,04 / 1,56	0,48 / 5,84	2,84 / 57,68	— / —	1,67 / 101,90	0,15 / 5,32	1,07 / 51,39	2,89 / 158,61	216,29
10—20	0,11 / 2,20	3,91 / 89,92	0,04 / 1,56	0,05 / 0,61	4,11 / 94,29	— / —	2,97 / 181,22	0,19 / 6,74	1,00 / 48,03	4,16 / 235,99	330,28
20—30	0,07 / 1,40	2,91 / 66,92	0,03 / 0,17	0,06 / 0,73	3,07 / 70,22	0,42 / 12,60	1,76 / 107,39	0,20 / 7,09	0,78 / 37,46	3,16 / 164,54	234,76
30—50	0,07 / 1,40	2,57 / 59,10	0,03 / 1,17	0,11 / 1,34	2,78 / 63,01	0,19 / 5,70	1,89 / 115,32	0,17 / 6,03	0,53 / 25,46	2,78 / 152,51	215,52
50—70	0,07 / 1,40	2,26 / 51,97	0,03 / 1,17	0,01 / 0,12	2,37 / 54,66	— / —	1,56 / 95,19	0,25 / 8,86	0,44 / 21,13	2,25 / 125,18	179,84
70—90	0,07 / 1,40	1,73 / 39,78	0,02 / 0,78	0,01 / 0,12	1,83 / 42,08	0,44 / 13,20	0,95 / 57,97	0,25 / 8,86	0,30 / 14,41	1,94 / 94,44	136,52

surpassing 10 mm, produced a relatively deep-lying subsoil waterlevel. This was changed only by the major precipitation of May-end and by the inundating irrigations; as a consequence of these, the soilwater level increased from 84 to 48 and later 42 cm, and decreased only in the Autumn period, in middle of October, to 90 cm again (Fig. 1).

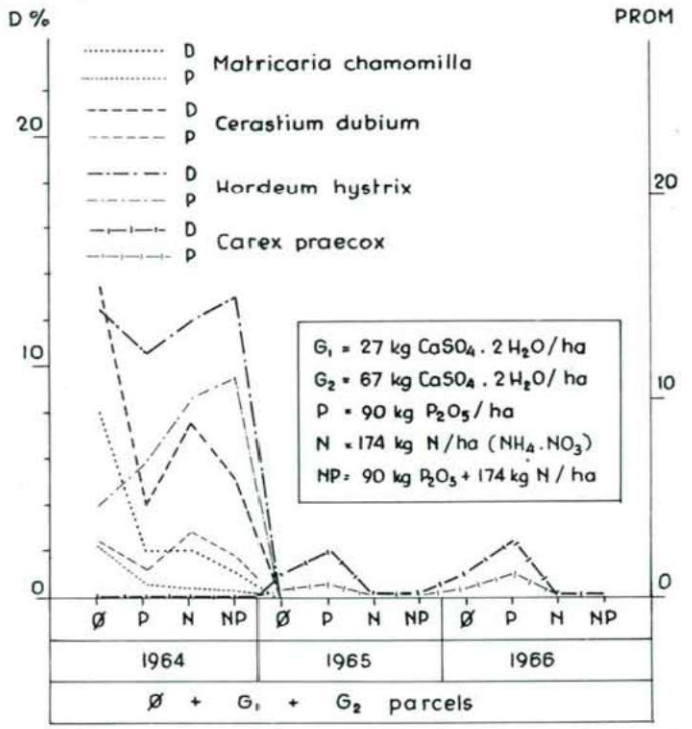


Fig. 3. Change of the dominance percentage and of the amount of surface production of some resettled species, as a result of different fertilizer doses.

Although the most roots of the meadow plant species in alkali soils take place in the upper 10 cm (HARMATI 1959), the high subsoil waters have a definitely positive effect on the development of the plant associations of our plots as well as under other, glycophilous cropland conditions (KLAPP 1954). That positive effect appears first of all in influencing the amount of soil moisture in a positive direction. This effect was examined by VAN'T WOULT and BESSEL (1955) in connection with the fertility of *Trifolium*. DANCAU (1962) reports some data connected with the water household of the plant species of meadows.

(2) S o i l. For clearing up what a soilchemical transforming effect the plastering may produce in the second vegetation year, the patterns of the different soil profiles were subjected to detailed laboratory examinations. According to the analysis of the water extracts, in Na_2CO_3 the Na ions don't exceed the 4 mg equivalent counted on 100 g soil in the unreclaimed plots in an accumulation level near the soil. In the upper 10 cm, woven by roots the most intensively, the

conditions are more propitious. Influenced by gypsum treatment, the amount of Na ions decreased therefore 1/3 in the layers of the upper surface, in a major dose 1/2.

The influence of the changeable Na on the species *Trifolium* and *Medicago* was examined by BERNSTEIN and PEARSON (1956); that of soil reaction and fertilizer supply on the meadow associations by BOECKER (1954). For the determination of Na-ions bound in the absorption complex we have used HERKE's method of determination instead of MEHLICH's routine procedure. Accordingly there was a major amelioration first of all in case of higher dose gypsum administration. Further details are given in Table IV. The connections between plant associations and soil conditions of natural meadows of glycophylous type are known on the basis of the examinations of BOER (1958) and MARSCHALL (1958). The alkaline differences between our plots reclaimed in different degrees, however, did not cause any major differences in the species combinations in the first two vegetation years if the inundation irrigation and proper fertilizers were ensured.

(3) V e g e t a t i o n. In the second vegetation year the experimental plots showed a considerable change concerning their species combination. It is obvious that the ephemerous halophytic *Puccinellietalia* elements, multiplied as a result of the breaking, fell fully into the background, while some sown species like *Arrhenatherum elatius* suffered a larger decrease, and other ones like *Poa angustifolia, Festuca pratensis* gained uniformly, and *Lolium perenne* suddenly, ground.

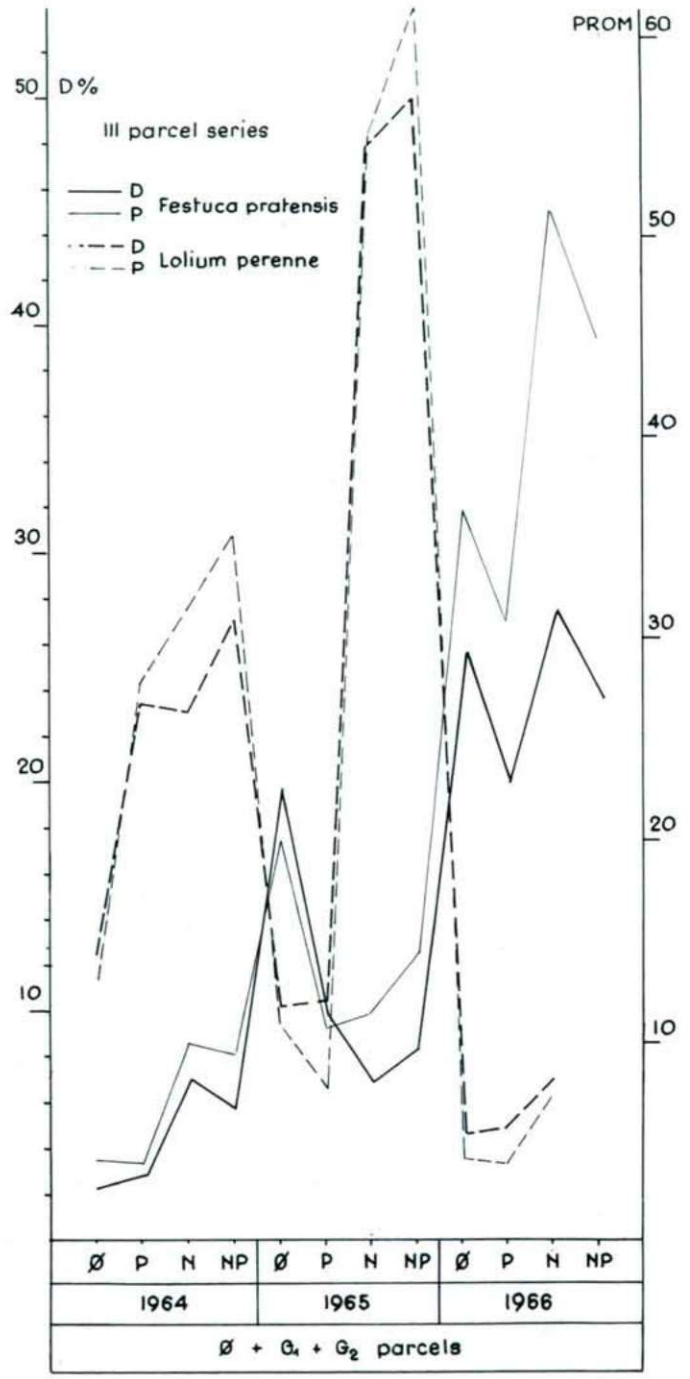
(a) The changes of plots of the formed meadow associations in respect of the grass level may be summarized as follows: on plots getting no fertilizer doses the groups of soil elements behaved differently on the basis of the means of the quadruple repetitions. The quantitative outset of *Lolium perenne* that could not get suitable fertilizers, first of all N, did not reach 10, being thus lower than it was in control plots of the beginning year. This may be attributed mainly to the increasing spread of *Festuca pratensis.*

Also some *Molinietalia* elements gain a considerable ground; the sudden advance of *Trifolium hybridum* observed in control conditions is especially remarkable; although the theree sown *Papilionaceae* ran to 21 per cent of the total seed used for sowing and the *Trifolium hybridum* only 2 per cent, its covering value being between 2 to 12 per cent. At the same time *Lotus corniculatus* has only a D—value of 0,5 to 2 per cent, and *Medicago sativa* occurs possibly only thread by thread.

The ecotype of bastard trefoil [Swedish clover] used by us is, unfortunately, not resistant enough to *Erysiphe* and its foliage is highly damaged especially in the Indian summer period.

(b) As a result of phosphorus treatment, a great number of the settled grass species have demonstrated a considerable recession, striking the eve particulary in case of *Festuca pratensis. Agrostis alba, Poa angustifolia;* in other cases, e. g., at *Lolium perenne,* the change is unimportant. It is even more obvious that also *Trifolium hybridum* demanding P lost ground (Fig. 4). At the same time, from

Fig. 4. Values of the coverage of *Festuca pratensis* and *Lolium perenne* (D) and of th amount of their surface production (P). PROM = ASP.


PROM
D %
III parcel series
D
P Festuca pratensis
D
P Lolium perenne
50
40
30
20
10
60
50
40
30
20
10
Ø P N NP Ø P N NP Ø P N NP
1964
1965
1966
Ø + G1 + G2 parcels

the introduced species, the *Carex praecox* occurs which, forming a dense rhyzoid-system, may be considered as a *Festuco-Brometea* species. Although its dominance value is low, its root concurrence plays a considerable role in forming the species combinations of the phosphorus-treated plots. The problem of competitions between the plant substances is treated of by BAEUMMER, BORNKAMM (1963); SPEIDEL (1966), as well.

The closing values of their grass levels are on a rather low degree. *Festuca pratensis,* dominant in the upper level, ensures a closing value of 6—12 per cent. The situation is better concerning the middle level where the values of the two resettled grass species, *Puccinellia limosa* and *Agrostis alba,* are raised by the setled *Poa angustifolia, Lolium perenne* and *Trifolium hybridum,* thus the value of closing of this level is 30—35 per cent or so. The lower grass level was represented by *Carex praecox* and the pushed off *Lotus corniculatus* with a rather low closing value, as the therophytic *Puccinellietalia* elements had disappeared and so did among them the *Hordeum hystrix* so widely spread in the first vegetation period.

(c) In the nitrogen-treated plots there took place highly important changes in covering and quantitative output as compared with those observed in the first vegetation year. There is remarkable first of all the nitrogen utilization of *Lolium perenne* whose quantitative output values have almost redoubled as compared to those of the last year. As influenced by an increasing concurrence, it ousted considerably the more and more efficient *Festuca pratensis* which accomodated very much to the dominant cropland conditions, together with the *Agrostis alba* of similar demand on a cropland. Opposite to it only the *Alopecurus pratensis* and *Poa angustifolia* proved to be competitive. The effect of fertilizers on *Lolium perenne* was, anyway, treated of by BERG (1962), and the association programs of *Lolium* species by VINCEFFY and co-workers (1954).

While the *Agrostion* species maintain their leadership after the spread of *Alopecurus pratensis,* the *Molinietalia,* resp. *Arrhenatheretea* species show a major recession in the Spring aspect of the second vegetation year. The *Papilionaceae* belonging here can hardly be found, mostly thread by thread.

In the upper grass level of these plots the leadership belongs already to *Alopecurus pratensis,* and beside it *Beckmannia eruciformis, Festuca pratensis, Bromus inermis* and *Arrhenatherum elatius* participate with their low covering in the closing degree of about 25—35 per cent only in a slight extent. In the middle level the closing is but of 65—78 per cent as a consequence of the covering percentage of *Poa angustifolia* and *Puccinellia* which are present apart from the mass presence of *Lolium.* Owing to the overshadowing of such a high degree, a lower grass level could not be formed in a continuous form.

(d) The double fertilizer doses of P and N have not brought about any considerable change. The covering percentage of *Lolium perenne* kept on rising. In the relation of grass level a situation similar to the plots with solo Nitrogen could be observed.

(e) In the Autumn aspect the sudden advance of *Trifolium hybridum* in the control and phosphorus-treated plots is the most obvious; it took up in control plots five times, in P-treated ones three times as large areas as in similar periods of the year before. This enormous increase is, however, not connected to the

original seed. *Trifolium hybridum* granulating fast after the June mowing attains earlier the state of a recent flowering than other components and its seed-ripening till the period of second mowing is as well ensured as the possibility of its further growth. Although these possibilities are given also in case of *Lotus corniculatus*, its covering percentage has not surpassed 8 per cent even in P-trea-

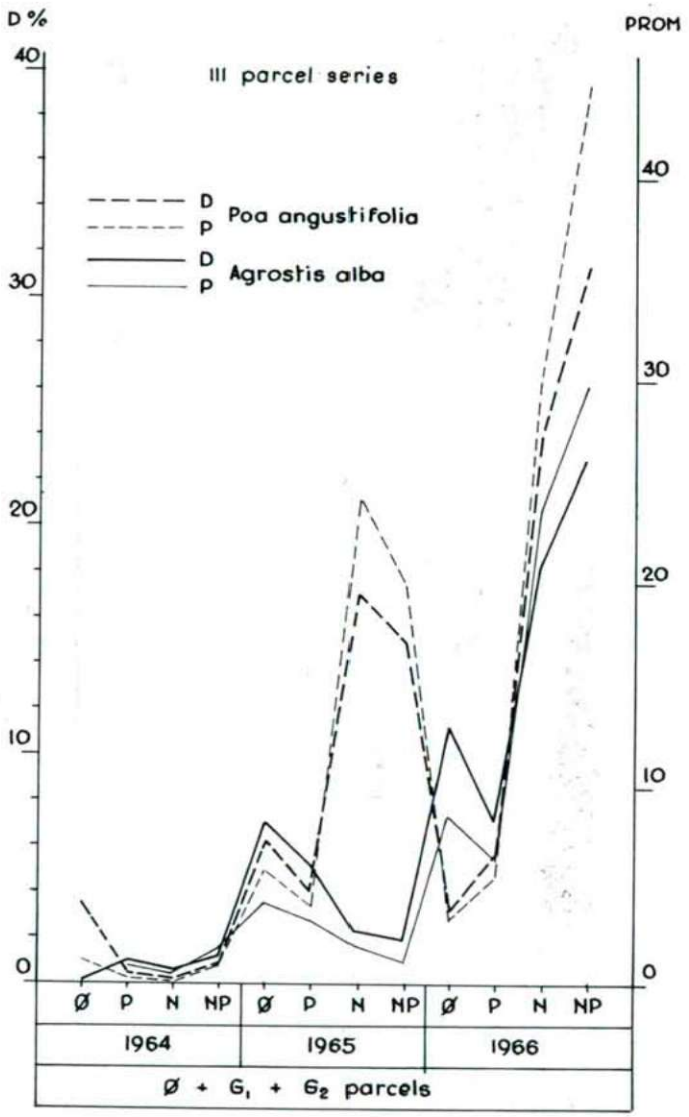


Fig. 5. Change of D and P values (P = PROM = ASP) of the *Poa angustifolia* and *Agrostis alba* during the period of examination.

ted plots. From that we can conclude the obviously good adaptability of *Trifolium hybridum* in a solonchak-solonetz soil of weakly saline surface soil of Danube valley under hygrophilous conditions. At the same time that species occured in the alkaline moorland of Danube valley not more than sporadically, (Fig. 6).

The Autumn aspect of the so produced meadow associations hadn't a major upper and lower grass level even at the end of the second vegetation year. All the more closed is the middle level, the dominant species being also here, the *Lolium perenne*. Its dominance values are, however, often approached by *Poa angustifolia*. That may be considered as a sign of the change in the dominating position of the two species. In P-treated plots the degree of this level-closing reaches, with *Papilionaceae* together, 80—90 per cent.

Results of the third vegetation year.

In the composition of the meadow associations settled in our experimental plots some considerable changes took place in the year 1966.

(1) As to the climate, our spring period continues being rainy and rather cool, in contrast to the accustomed lowland climate. In the air layer of the grass level, the maximum of air temperature did not surpass 30 C° even in June, apart from one exception. The more uniform distribution of precipitation was completed by an inundating-irrigation employed twice in May. Even the level of subsoil water of this period is high; it is 30 cm or so on the average, sinking under 60 cm only in May (Fig 1).

As influenced by the favourable meteorological and the consequent soil ecologic events at our meadow associations, a change took place to the advantage of the glycophyta in the area of the settled species. Thus the *Puccinellia* which had, with lesser fluctuations, a 6—10 per cent covering value in the former years, has now suffered a considerable loss of area in relation to *Agrostis alba* similarly resettled. At any rate, its lower dominance values may also be attributed to the cool Spring climate not favourable for it. As a consequence of the undisturbed meadow development, also the therophytic *Puccinellietalia* elements were fully driven back.

(a) In our plots without fertilizers, on the basis of the species combination of earlier years, we have to emphasize two important changes: the considerable recession of *Lolium perenne* and the parallel advance of *Festuca pratensis* in the control plots, resp. the advantage of *Agrostis alba* opposite to *Poa angustifolia*. *Agrostis* figured in the control plots in a similar way in the similar periods of all the three years, the inference being that it has a rather weak competitiveness. *Trifolium hybridum* continues gaining place even in control plots after seed-dropping.

(b) The favourable influence of the phosphorus dose is again repeated at the most kinds of grass. The decline of *Festuca pratensis* and *Agrostis alba*, similar to that in the former year, concerning both covering percentage and quantitative output, is particularly remarkable. It is the most favourable from the point of view of *Trifolium hybridum* and *Lotus corniculatus* belonging to *Molinietalia*

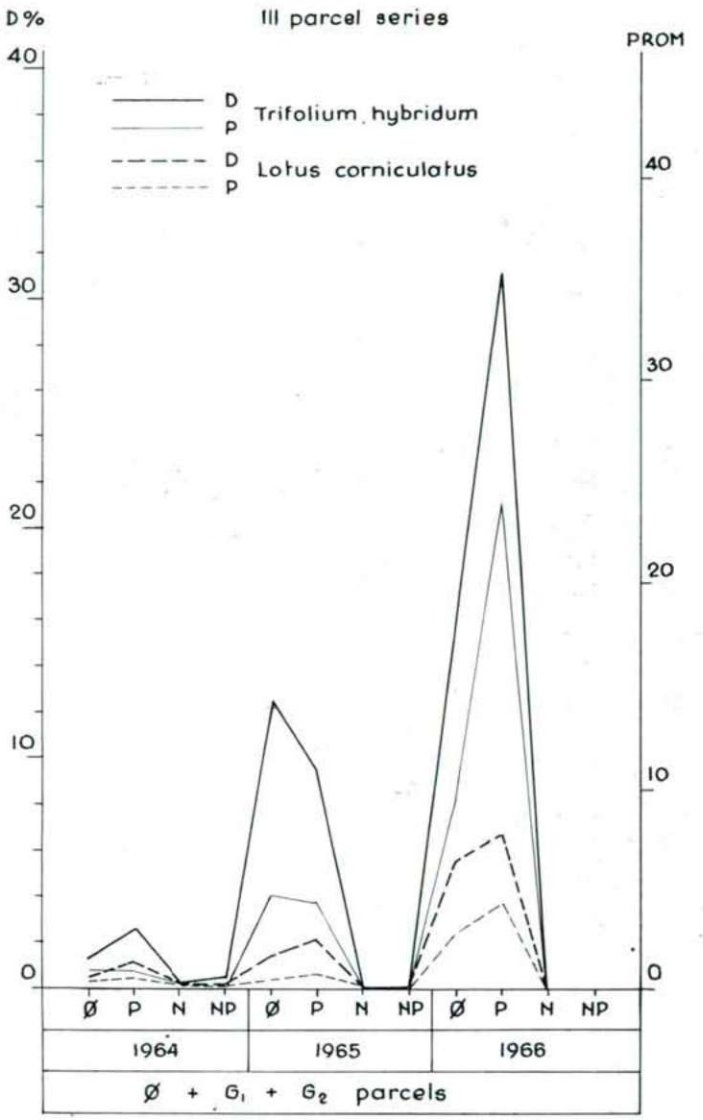


Fig. 6. Changes of D and P values (P = PROM = ASP) of the two *Papilionaceae* species of the association in the culture meadow.

elements whose spreading is still rising, for the time being, in direct ratio to the number of years; the covering percentage of *Trifolium* surpasses 30 per cent, and its quantitative output 35 per cent. On the other hand, *Medicago sativa* disappeared fully even from the P-treated plots.

The P-effect is definitely advantageous to the development of *Carex praecox*, as well, apart from the mentioned *Papilionaceae*, in our experimental tables, without enabling, anyway, a major spread of the *Festuco—Brometea* species in the third year.

(c) In the Nitrogen-treated plots, a major decline of *Lolium perenne* catches the eye, without being able to be prevented even by N-administration. At the same time, we have to reemphasize the sudden increase of the quantitative output of *Festuca pratensis* as compared with the covering percentage. The same may be ascertained also in case of *Poa angustifolia* which employs the N-fertilizer extremely profitably. Its multiplication is obviously high: 25—35 per cent, first of all in the Nitrogen-treated plots (Tab. VI).

It is characteristic of the level of these plots that in their upper grass level *Festuca pratensis* keeps on dominating; also *Alopecurus pratensis* is rather considerable here and there: its increase has, however, not even approach that observed in case of the cultivated meadows set up in the solonetz soils beyond the Tisza (BODROGKÖZY 1962, 1965).

The closing degree of the middle grass level is the highest first of all as a consequence of the prevalence of *Agrostis alba*, resp. *Poa angustifolia*. The opposite formation of the power relations may be explained mainly by the discontinuance of the concurring influence of *Lolium perenne*, measurable by the change of the covering percentage of *Agrostis alba*, as well.

Problem of succession of the ancient covering vegetation. On the plots there could not be observed even an initial state of the original species combinations of the *Lepidio-Puccinellietum asteretosum* which was dominant at the beginning of experiments; later, however, it was broken. As influenced by irrigation and Nitrogen doses, the cenoses of plots may be considered as a cultivated subassociation of *Astero-Agrostetum Poa angustifolia*, as a consequence of the change of cropland conditions, whether or not some fertilizers were applied. In its species combinations the settled and resettled species have the leadership. Even today, these can be observed everywhere in the weakly saline solonchak solonetz soil settled on sand in the moorlands between Danube and Tisza, having a better water supply. Both from point of view of level proportion and from that of output they are standing on a more developed degree than the *Astero-Agrostetum* substances of the croplands of Danube plains with a larger backwater content (Tab. VII).

(d) The double-treated (P+N) plots yielded the highest total quantitative output of the third vegetation year (Fig. V). The influence of the double fertilizer doses can be observed first of all at *Poa angustifolia*, *Festuca pratensis*, however, showed some recession. This may be explained probably not only by the effect of the power relations of the concurrence but by the for it unfavourable influence of phosphorus, as well, taking place not only in case of the solo P-treatment but also in that of a double treatment.

Their species number, like in the solo-N-treated plots, decreases also here, and their cenoses consist of hardly 6—8 species. Apart from the sown grass species as well from both resettled ones, other settled species can hardly be observed, at most only thread by thread, even in three years after the settlement.

Connection between the total output and the dry hayweight. The spatial distribution of the overground mass of plant substances is such that a great

part of the vegetative mass of grasses is concentrated nearer the soil than at the *Papilionaceae*. Thus in the species combinations of *Gramineae* and *Papilionaceae* species a more uniform distribution of output may take place, and their output is greater than that of pure substances (ALEKSEENKO 1958). The sum of the output calculated for species on the basis of dominance values and height averages may only be considered as a real value if it doesn't differ considerably from the values obtained by measurements (dry-hay). For control and easier perspicuity the first mowing dry hay-weight of the single vegetation years are charted according to plots. Drawing the curve of mean values, we have proceeded similarly with the values of the total quantitative output, as well. The connections observed in the Spring aspect of the vegetation year 1966 are demonstrated in Fig. 7.

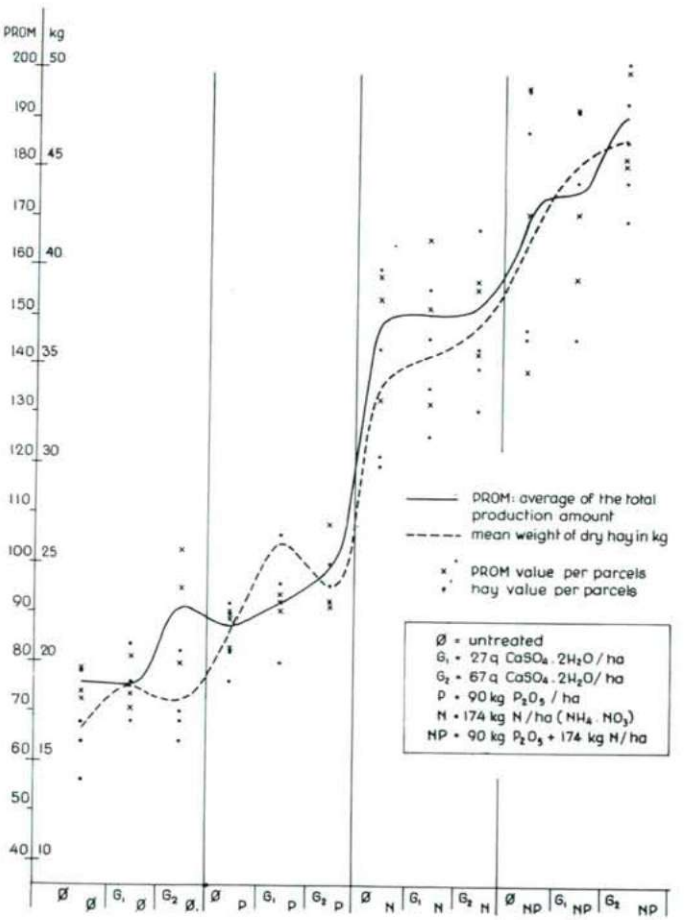


Fig. 7. Interconnection of changes of the amount of surface production (PROM = ASP) of the settled-meadow association and those of the air-dry hay-weight per parcels in June 1966.

TABLE V

Unfertilized parcels (June 3rd 1966)

Fertilizer and its amount:	O				$CaSO_4 \cdot 2\ H_2O$							
					27 q/ha				67 q/ha			
Number of parcel series:	I.		III.		I.		III.		I.		III.	
Number of all species:	13		12		12		11		12		8	
Dominance and production amount:	%	P	%	P	%	P	%	P	%	P	%	P
Puccinellion species:												
H Puccinellia limosa	4	3,8	4	3,7	4	4,0	5	5,0	4	4,0	4	4,0
Festucion pseudovinae species:												
H Scorzonera cana	0,2	0,1	0.5	0,4	0,5	0,3	·	·	1	0,2	·	·
Agrostion species:												
H Festuca pratensis	30	34,5	20	26,1	25	33,5	20	26,8	35	48,3	35	48,3
H Alopecurus pratensis	2	2,4	1	1,3	1	1,2	1	1,4	1	1,2	·	·
H Agrostis alba	3	2,3	8	8,0	5	3,4	10	7,7	8	6,4	10	6,3
H Ranunculus sardous	0,5	0,2	2	0,8	·	·	1	0,4	·	·	1	0,5
Molinietalia and Arrhenatheretea species:												
H Phleum pratense	·	·	2	1,4	·	·	2	1,3	·	·	2	1,1
H Arrhenatherum elatius	2	1,3	·	·	1	0,7	·	·	1	0,7	·	·
H Trifolium hybridum	15	9,2	16	9,8	20	13,4	14	8,0	20	11,1	12	7,1
H Trifolium repens	·	·	·	·	0,5	0,2	·	·	·	·	3	2,5
H Lotus corniculatus	8	4,0	4	1,6	10	5,3	5	1,7	10	5,4	2	0,6
H Taraxacum officinale	12	0,3	·	·	1	0,1	·	·	1	0,1	·	·
Bromion and Festuco-Brometea species:												
G Carex stenophylla	2	0,3	3	1,0	1	0,1	1	0,2	1	0,2	·	·
H Poa angustifolia	10	9.6	8	7,3	6	5,5	0,5	0,4	6	5,3	·	·
Association-indifferent species:												
H Lolium perenne	·	·	7	5,6	·	·	12	9,6	1	0,6	·	·
H Mentha pulegium	1	0.1	·	·	·	·	·	·	·	·	·	·

TABLE VI

P_2O_5 (90 kg ha) treated parcels June 3rd 1966

Fertilizer and its amount:	O				$CaSO_4 \cdot 2\ H_2O$							
					27 q/ha				67 q/ha			
Number of parcel series:	I.		III.		I.		III.		I.		III.	
Number of all species:	10		11		13		10		11		10	
Dominance and production amount:	%	P	%	P	%	P	%	P	%	P	%	P
Puccinellion species:												
H Puccinellia limosa	5	4,4	5	5,1	4	3,2	5	4,1	4	3,7	3	2,7
Festucion pseudovinae species:												
H Scorzonera cana	·	·	0,2	0,1	0,5	0,4	·	·	0,2	0,1	·	·
Agrostion species:												
H Festuca pratensis	23	33,8	20	30,7	20	24,9	20	28,8	20	27,6	25	38,3
H Alopecurus pratensis	·	·	·	·	3	4,6	·	·	·	·	·	·
H Agrostis alba	6	4,8	7	6,3	7	6,4	8	7,0	6	4,4	8	7
H Ranunculus sardous	0,5	0,2	0,2	0,0	0,5	0,2	1	0,5	0,5	0,3	0,5	2,6
Molinietalia and Arrhenatheretea species:												
H Arrhenatherum elatius												
H Phleum pratense	4	2,4	4	2,3	4	3,8	3	2,4	4	2,4	4	3,4
H Trifolium hybridum	18	13,8	30	23,0	23	16,0	30	21,8	35	25,5	33	26,6
H Trifolium repens	·	·	·	·	0,5	0,2	·	·	·	·	·	·
H Lotus corniculatus	8	4,3	4	2,1	15	9,0	4	2,5	7	4,9	5	2,4
H Taraxacum officinale	·	·	·	·	0,5	0,0	·	·	·	·	·	·
Bromion and Festuco-Brometea species:												
H Poa angustifolia	5	4,0	5	4,3	10	8,0	5	5,0	6	5,3	5	5,0
G Carex stenophylla	1	0,2	3	1,4	·	·	2	0,3	1	0,2	6	2,4
Association-indifferent species:												
H Lolium perenne	3	2,4	4	2,6	2	1,6	6	4,3	6	3,7	6	5,7

TABLE VII

$NH_4 \cdot NO_3$ (174 kg N/ha) treated parcels June 3rd 1966.

Fertilizer and its amount:	O				$CaSO_4 \cdot 2\ H_2O$							
					27 q/ha				67 q/ha			
Number of parcel series:	I.		III.		I.		III.		I.		III.	
Number of all species:	6		9		6		8		7		8	
Dominance and production amount:	%	P	%	P	%	P	%	P	%	P	%	P
Puccinellion species:												
H Puccinellia limosa	.	.	8	11,9	.	.	6	7,3	.	.	3	3,4
Festucion pseudovinae species:												
H Scorzonera cana	0,3	0,2	1	1,0	.	.	.	.	1	0,4	0,5	0,4
Agrostion species:												
H Festuca pratensis	20	34,5	18	29,3	22	40,0	20	36,4	30	57,5	27	44,0
H Alopecurus pratensis	.	.	8	14,1	2	3,7	6	6,9	4	7,2	4	6,9
H Agrostis alba	30	40,8	30	40,2	30	39,1	18	22,4	14	19,3	15	18,7
Molinietalia and Arrhenatheretea species:												
H Arrhenatherum elatius												
H Phleum pratense	9	9,4	3	2,9	6	8,2	4	4,6	10	10,9	8	10,0
H Trifolium hybridum	.	.	.	.	.	.	0,5	0,0	.	.	.	.
H Trifolium repens												
H Taraxacum officinale			0,5	0,0	.	.	.	.	.	.	.	.
Bromion and Festuco-Brometea species:												
H Poa angustifolia	35	41,6	25	27,8	35	44,3	35	43,6	25	29,7	30	37,4
G Carex stenophylla	.	.	.	.	.	.	.	.	.	.	.	.
Association-indifferent species:												
H Lolium perenne	5	5,1	3	3,4	5	4,6	4	3,5	6	7,2	5	5,7

Summary

A considerable part of the Pannonicum halophilous vegetation can be found as developed in the eroded solonchak-solonetz soils in the Danube valley. These alkaline meadow associations which belong at most to the different subassociations of *Lepidio-Puccinellietum* are of low species number and are standing on a low degree of succession, depending upon the quantity of sodiumcarbonate-hydrocarbonate. And they are caused also, to a high degree, by the fertilizer deficit of the soil devastating effect of the inland-water erosion. For clearing up this question, in the Autumn of 1963 we arranged an experimental series, repeated two times, under irrigated conditions, in the area of Kiskunlacháza, carrying out also soil-amelioration by dressing the soil with sulphate of lime, apart from phosphorous, nitrogen, and their combination. After we had broken the original *Lepidio-Puccinellietum asteretosum* of the experimental plots, relying on the selective effect of sodium salts, we used a seed mixture composed of several species. In three years we have obtained the following major results:

(1) In spite of the weakly saline solonchak-solonetz soil, a great part of species sprouted, producing multilayer meadow cenoses in the midst of a strong struggle for life. For measuring the different influences, we took for a basis the changes of dominance relations and quantitative output according to species, apart from keeping in mind the changes of air temperature, precipitation, and irrigation conditions on the grass level, as well as those of the subsoil-water level.

(2) In the period after the breaking, soma ephemerous halophytae, first of all *Puccinellietalia* elements like *Matricaria chamomilla v. salina, Cerastium dubium, Hordeum hystrix* appear temporarily en masse.

(3) Az a result of N-doses, at first *Lolium perenne* took the lead, then step by step *Festuca pratensis, Poa angustifolia* and the ancient, resettled *Puccinellia limosa* and *Agrostis alba* prevailed.

(4) As a result of phosphorous, the *Molinietalia* species, represented by the *Papilionaceae: Trifolium hybridum* and *Lotus corniculatus* began more and more to prevail. Their prevalence increased also in the way of seed-spreading. The P-doses have a consequently negative influence on some grass species, particularly on *Festuca pratensis,* even in case of a double treatment (P+N).

(5) In the years after the breaking, the succession of the original *Lepidio-Puccinellietum asteretosum* has become extremely slower; and even it changes after being influenced by the soil amelioration through a systematic irrigation (whose influence on vegetation could not be demonstrated in the first three years) and by the administration of fertilizers, becoming similar to the *Astero—Agrostetum* species combination of the moorlands between Danube and Tisza.

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EFFECT OF ECOLOGICAL FACTORS ON THE LEAF EPIDERMIS OF SPECIES SOLANUM

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The tissue structure of leaf epidermis is examined by several papers.

According to the majority of scientists, some of the tissue elements of epidermis maintain conservatively the features of species being thus useful as a systematic mark at diagnostificating the single species.

ZÖRNIG and WEISS (1925) consider the relation of the epidermis cells and the palisade parenchyma cells below them to be constant; WALLIS (1946), on the other hand, mentions the vein-islet number of leaf, the stomatal ratio and stomatal index as marks of diagnostical importance.

SÁRKÁNY and RIEDEL (1951) emphasize length and width of the guard cells of stomata, resp. their quotient, the stomatal size; and MARÓTI (1965), in the course of his investigations on the leaves of Pteridophytes, found the ratio of perimeter per surface of the epidermis cells as well as the percentage distribution of the subsidiary cells of stomata to be constant.

The above-mentioned qualities are quantitatively measurable characteristics of the epidermis. From the formal, qualitative qualities of the epidermis there are known the shape of epidermis cells, the microstructure of their cell-walls, the forms of trichomes, the way of the origin of stomata, the quality of silica-cells as marks for determining the species (LINSBAUER, 1930; SÁRKÁNY and FILLÓ, 1951; MARÓTI, 1965).

Recently we have found more and more allusions in the scientific literature to the fact that also the epidermis of plants reacts sensitively to the environmental effects from outside, and the several ecological factors (light, temperature, soil conditions) can change the stucture of the skin-tissue-system, as well (SINNOTT, 1960; SIMON—WOLCSÁNSZKY, 1964; SHANKS, 1965).

The present paper is treating of the examination of the effect of the external environment on the leaf epidermis.

In the course of our experiments we have wanted to get an explanation whether or not the quantitative epidermal features of the specimens of a species *Solanum*, developed in different surface soil relations, show up differences significant enough to unfit the epidermal quality in question for diagnosticating the species or to make questionable its reliability.

There were measured the following epidermis values: the relative number of stomata, stomatal index, length and width of stomata and their size (stomatal

length per width). For valuation of results the mathematic-statistical method was applied, highly increasing in that way the objectivity and exactness of the valuation of our results.

Materials and methods

Some species of *genus Solanum*, showing up a great abundance of species, have been used for being examined. *S. dulcamara* L. and *S. nigrum* L. are growing wildly in Hungary, while *S. laciniatum Ait.*, *S. giganteum* JACQ. *S .sodomaeum* L. are cultivated experimentally because of their great steroid-alkaloid-glycosid content.

For examining the plant-ecological effects of the different soil relations, specimens of *S. dulcamara* L., of *S. nigrum* L., and of *S. laciniatum* AIT., grown in five-five different soils were gathered in. The croplands were chosen in the way that two-two specimens were ingathered at all the three species from places of an extreme soil.

There were always selected plan specimens in flower and the leaf samples were taken from them. From every soil the leaf samples were taken from the middle leaf region of the stalks of three-three plant specimens. The gathered leaves were fixed in JUEL'S mixture, boiled in SCHULZE'S macerating solution, and after rinsing they were stained by EHRLICH'S haematoxylin-vesuvin double staining. The so prepared epidermis preparations from the upper and lower surfaces were conserved in glycerin-gelatin.

For determining the number of epidermal cells and stomata, the preparations were projeced from a microscope placed under the drawing table. The length and width of stomata were measured by an ocularmicrometer. At every preparation we measured 30—30 fields of sight, i. e., 90—90 data in every cropland. For valuating the results in biometric way, we prepared the curve of distribution of the several groups of data. These curves showed forms being very similar to GAUSS'S curves; the data of our measures have, therefore, been of a very normal dispersion.

The valuation of results was carried out by variancy analysis. At preparing the basic tables of variancy analysis, we formed three-three lines and five-five columns.

In the lines the arithmatic means of the 30—30 measurement-data of three-three preparations made from specimens grown in the same cropland were registered.

The five-five columns contained epidermis data corresponding to the plants of the five different soils.

Thus we have obtained basic tables with fifteen headings.

Then the variancy tables were constructed and the corresponding calculations carried out.

In the course of the significancy examinations, the validity of our conclusions from the numerical data of our examinations was controlled by F- (FISHER) and t- (STUDENT) tests.

It was settled by F-test whether or not the values of the numerical differences between the different soil means are greater than the means of epidermis values of the specimens of the same soil on a $P=5$ p. c. probality level.

We calculated by t-test the differences inside which the values may only be attributed to test failures caused by the differences of sample selection.

The SD_5 per cent values are made known at the description of results.

Results

(1) Determination of the site of sampling

Before examining the effects of the environmental factors, we considered to be necessary to determine the site of the epidermal excoriation. We know ZALENSKY'S statement (1904) aceording to which the number of stomata is gradually growing as we advance from the lower part of stalk to the top of that. Accordingly, it is not indifferent from which nodules of the stalk of a plant the leaf samples are obtained. Some researchers found differences between the stomatal

numbers of the basis, middle, and apex of the leaf sheet even inside the single leaves. According to SLAVIK (1963), there are significant differences between the stomatal numbers of the basal and apical parts of the tobacco leaves. SIMON—WOLCSÁNSZKY (1964) has observed a decrease of the stomatal number characterized by a regressive straight, advancing from the basis to the apex of the leaf sheet of yellow corn.

For these examinations we have selected the *S. sodomaeum* L. of small stature and of small leaf size the. *S. laciniatum* AIT. of middle stature and of middle leaf size, and the *S. giganteum* JACQ. of tree-stature and of large leaves. Leaves were gathered in from the nodules of the lower, middle, and upper parts of three-three flowering specimens of all the three species. There were excoriations prepared from the lower surface of leaves at the basis, leaf middle, and apex of the leaves gathered in. On every preparation the number of stomata of a field of 60 sq. mm was valuated statistically. The results are demonstrated in Figs. 1/a and 1/b.

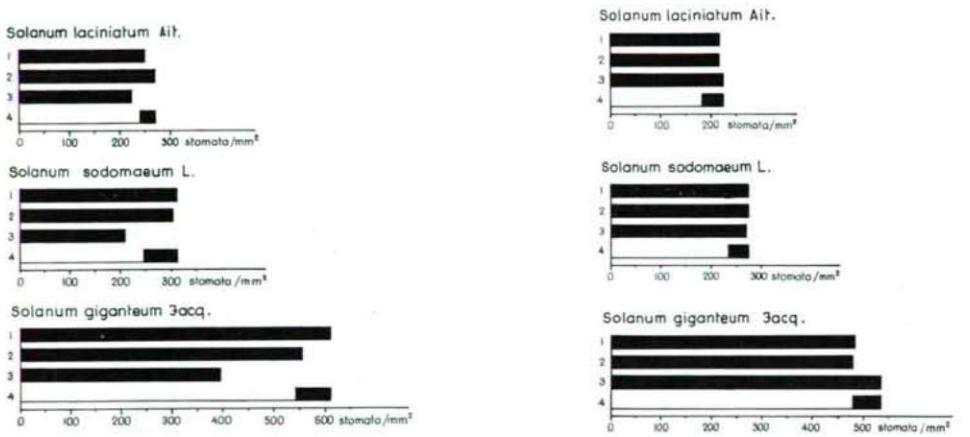

Fig. 1. a. Stomatal number of the leaves of stalk arising different heights (lower epidermal surface).
1 = upper leaf region;
2 = middle leaf region;
3 = lower leaf region;
4 = SD_1 p. c.

b. Somatal number of different parts of the leaf sheet (lower epidermal surface).
1 = leaf apex;
2 = leaf centre;
3 = leaf basis;
4 = SD_5 p. c.

It can be ascertained that:

(a) At the leaves arising from different heights of the stalks of species *Solanum,* the turn of the number of stomata follows roughly ZALENSKY's rule, *i. e.*, the stomatal number is the smallest at leaves in the lower part of stalk, going towards the apex, however, it grows gradually. This can be seen the most clearly

in the case of the *Solanum giganteum* JACQ. of tree stature, while in the case of the other two species only the stomatal number of leaf region differs (even on SD_1 per cent level) from the number of stomata of the other leaf regions. It is advisable to take a leaf sample from the middle part of stalk as the stomatal number of the middle leaf region gives the arithmetic mean. We have possibly to select leaves of identical position, node for every comparison.

(b) At the formation of stomatal number inside the leaf sheet it can be noticed that the difference between the single parts of leaves is but minimal. Only the stomatal number of the leaf basis of *Solanum giganteum* JACQ. with 25—30 cm life size differs significantly on SD_5 per cent. Regarding also the data of SLAVIK (1963) and SIMON—WOLCSÁNSZKY (1964), we may draw the conclusion that the differences between leaf parts are to be taken into consideration chiefly in the case of leaves of large size. In that case, too, it is the best to prepare the excoriation from the middle of leaf sheet. We consider as important to be noticed that the two sides of the main vessel of leaf are no reflections of each other in a mirror; it is therefore advisable to take samples from both sides of the main vessel.

(2) The effect of differring cropland conditions on the leaf epidermis

During the epidermis examination of the plants of *S. laciniatum* AIT., *S. dulcamara* L., and *S. nigrum* grown in five-five different croplands the greatest differences were observed concerning the change of the epidermal cell number. The number of epidermal cells shows up great differences at species developed in different conditions. It can be observed that we can find much more epidermal cells on the lower epidermal surface than on the upper surface. That is the cause of being very few stomata on the upper surface, on the lower surface, however, many ones. The stomata are of *Cruciferae*-type (METCALFE and CHALK, 1950), therefore every stomata being surrounded by three-four subsidiary cells. Thus it can be understood why these cells are smaller than the epidermal cells of the upper surface. The ratio of the cells of the lower epidermal surface per those of the upper epidermal surface is, however, constant in every cropland. The formation of the relative stomatal number, stomatal index, stomatal length and width, as well as that of the stomatal size in different croplands can be observed in Figs. 2, 3, 4, 5 and 6. In the column diagrams, the white columns are showing the data of the upper epidermal surface, the black columns those of the lower epidermal surface. In every figure also the SD_5 per cent values are given, these being the values of the greatest differences still acceptable.

Also the number of stomata belonging to the unit of a leaf surface demonstrates great differences. Projecting differences are, however, to be found but at the species developed in extreme conditions, the data of the three-four similar croplands are close to one another. This is connected with the finding that the plants react upon the effect of the outer circumstances first of all by the size of cells; that however, may not be a dicisive factor, as proved by the change of stomatal index.

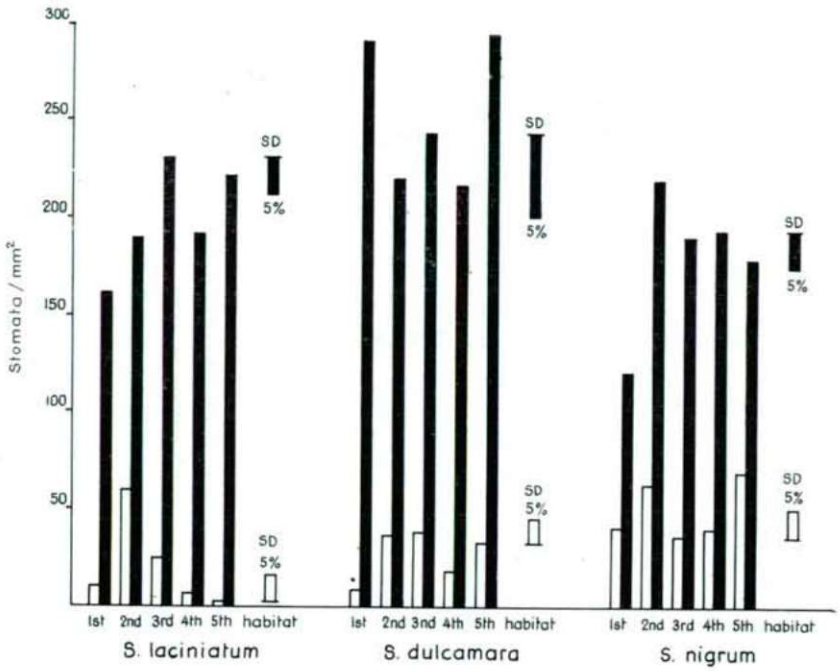

Fig. 2. Formation of the stomatal number at specimens grown in different croplands.

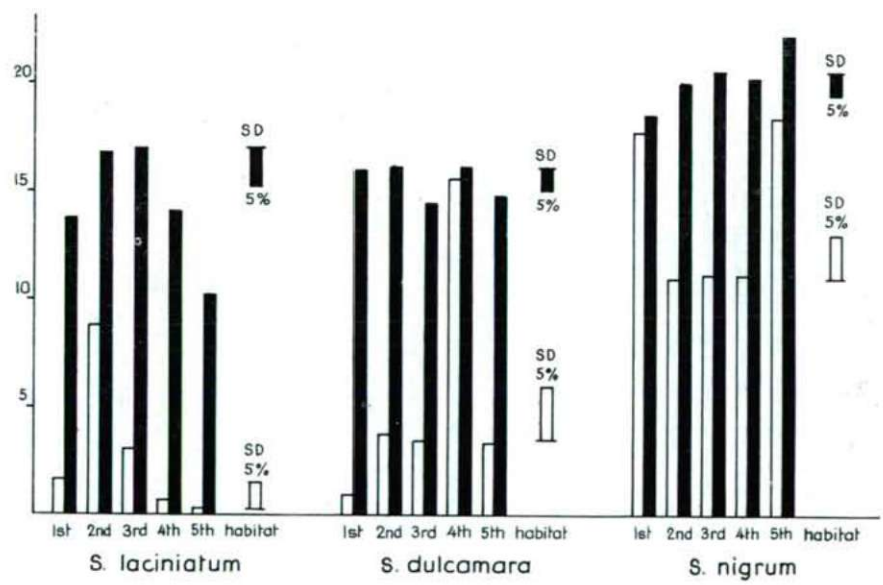

Fig. 3. Change of the stomatal index under the influence of environmental effects.

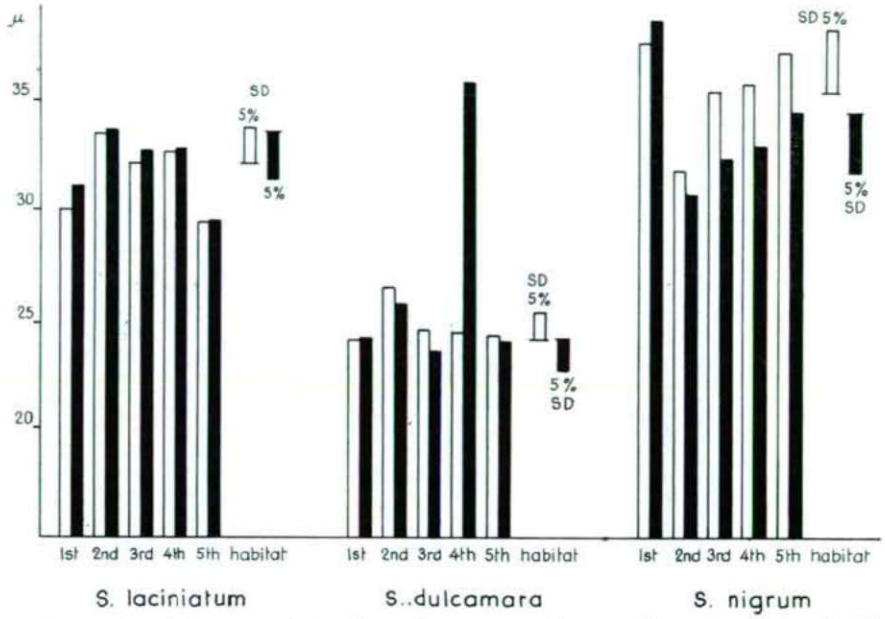

Fig. 4. Formation of the stomatal length on the upper and lower epidermal surfaces of *Solanum* leaves.

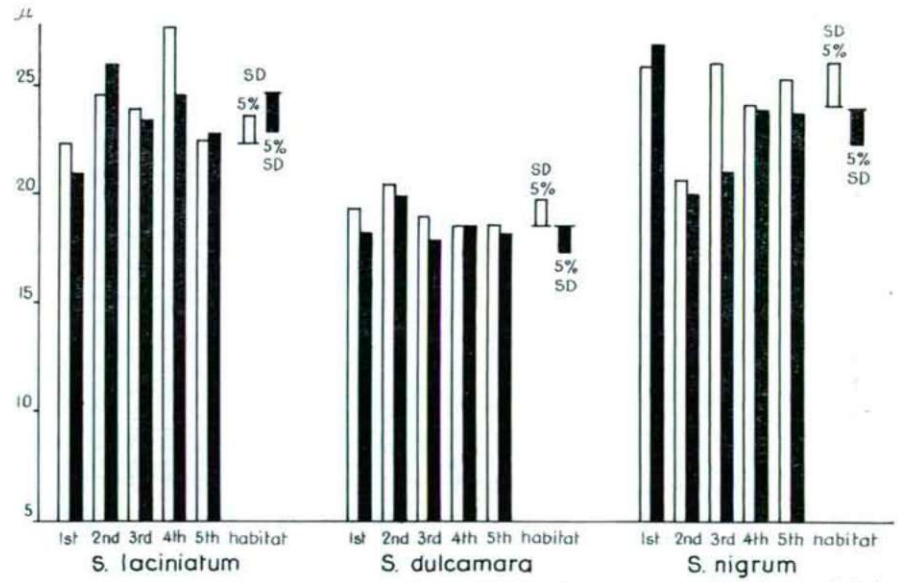

Fig. 5. Formation of the stomatal width on the upper and lower epidermal surfaces of *Solanum* leaves.

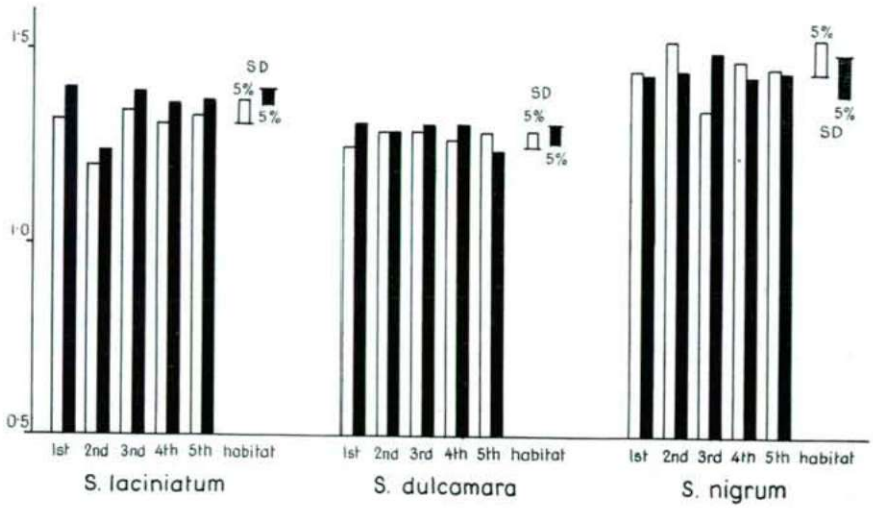

Fig. 6. Formation of the stomatal size on the upper and lower epidermal surfaces of *Solanum* leaves.

According to some authors, the ratio of epidermal cells and of stomata to each other is determined genetically. This means that even if the extent of stomata and epidermal cells changes, influenced by different ecological factors, their size increases or decreases proportionately, and therefore the stomatal index (stomatal number per number of epidermal cells plus stomatal number) cannot change.

It can be observed in Fig. 3. that the stomatal index shows differences similar to the stomatal number; between specimens developed in ecologic conditions similar to each other the difference is of lesser degree, the stomatal index of the leaves of individuals living under extreme condition differs, however, even on SD_1 p. c. level from that of other ones. The external circumstances exercise, therefore, a significant influence on the internal, genetical connections of the skin tissue.

Length and width of the guard cells of stomata change at the Solana living under different conditions in a smaller degree than the stomatal number and index; nevertheless, even these are not suitable to determine the single species with absolute certainty. It is obvious that even the guard cell data of the stomata of upper and lower surfaces are not agreeing thoroughly with one another; the difference of their sizes is, however, inside a probability level of 5. p. c.

The stomatal size (length per width of tomata) is a ratio, it gives a directing value concerning the shape of the guard cells of stomata.

It was indicated already by the values of F-test that there are no differences here between the values of the different soil data that could be valuated on SD_5 p. c. The stomatal size is, therefore, the most stable epidermal mark among the qualities examined until now.

Discussion

(1) It can be ascertained on the basis of valuation of the stomatal numbers measured in different regions of stalks and leaves that the middle of the leaf sheet obtained from the middle stalk part is suitable to be examined.

(2) The quantitative epidermis qualities of the species *Solanum* developed under different soil conditions show up differring changes if influenced by the environment. The stomatal size changes the least. The stomatal length and width, as well as the stomatal index and relative stomatal number undergo a more considerable change.

For diagnosticating the species, the stomatal size is the most suitable. The employment of the other epidermal marks is less reliable; it can be employed with succes only for species living under similar conditions. Genus *Solanum* is one of the genera with the highest species number, still developing. This is manifested in reacting to the effects of the external environment more sensitively as well, and is reflected also by the formation of some quantitative qualities of the leaf epidermis as emphasized above.

(3) At the analysis of the leaf epidermis on the basis of quantitative epidermal qualities we consider the employment of biometrical methods to be absolutely necessary.

Summary

We have examined the change of some epidermal qualities, measurable quantitatively, on the leaves of specimens of *Solanum laciniatum* Ait., *S. dulcamara* L., and *S. nigrum* L., grown in different croplands. It was ascertained that the external, ecological conditions had a considerable influence on the tissue structure of the leaf epidermis, making questionable the species-diagnostical value of the examined epidermal marks.

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DEVELOPMENT OF THE TMESOPSIDA AND PTEROPSIDA LEAVES AND HISTOGENESIS OF THE EPIDERMIS

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Introduction

The morphogenesis of leaves, the development and structure of epidermis and stomata reflect in many cases the degree of development and natural taxonomic place of the species and family. The problem of the histogenesis of epidermis has been in the centre of interest for about a hundred years. The first examinations concerned mainly the genesis of stomata. The first observations concerning the development of stomata are connected with names of OUDEMANS (1866), HILDEBRAND (1866), PRANTL (1872) and STRASBURGER (1867). OUDEMANS (1866) could not, as yet, form a decisive resolution in connection with the peculiar development of stomata of the *Anemia species,* therefore he thought on four possibilities of the development. HILDEBRAND (1866) has already examined the development of stomata of more ferns, being anxious to discern some types. He named his types after the species. These types cannot be generalized, they are mechanical in nature, without reflecting the development. STRASBURGER (1867) and PRANTL (1872) have tried to give the first rule of the genesis of stomata with a universal validity. On the basis of their investigations two main types may be distinguished:

The guard mother cell develops with a single division from the primary mother cell.

The cell, left over in the middle part of the cell after a series of divisions of different directions in the primary mother cell, becomes the guard mother cell.

It became, however, evident in the course of later examinations that a number of plant groups cannot be included in the main type considered to be of general validity. PRANTL (1872) and STRASBURGER (1867) included the *Pteridophytae* and *Gymnospermata* incorrectly uniformly in the first group as but a number of classes may have been included here. This formulation allows of several misunderstandings.

The conclusions concerning the development of the epidermis of the several species are durable and highly useful in the synthetizing work. From the many literary data author mention but those treating of the *Pteropsidae.* RAUTER

(1870) compared the stoma development of *Niphobulus linqua, Anemia fraxinifolia* and *Pteris longifolia.* PRANTL (1881) observed, in the course of his anatomic and taxonomic examination of *Schizaeaceae,* even the formation of stomata of the *Schizaea pennula.* Britton and TAYLOR (1909) treated also of the formation of leaves and development of stomata of the *Schizaea pusilla.*

In the examination of histogenesis and historical development of stomata considerable results have been achieved by FLORIN (1931, 1933), who has distinguished, in the course of his examinations of the epidermis of *Coniferales* and *Cordaitales,* three main types of the history of development of the genesis of stomata:

A) The primary mother cell (of guard cells) becomes immediately guard mother cell.

B) The guard mother cell is, after the formation of the dividing wall, adjacent to the primary mother cell.

C) By the prepared division of the primary mother cell the guard mother cell precedes the development of the adjacent cells.

FLORIN (1933) mentions only two types in his work dealing with *Cycadales* and *Bennettitales:*

A) Haplocheil type:

1. The primary mother cell of guard cells functions immediately as a mother cell of these cells. This guard mother cell is divited by a longitudinal wall into two guard cells.

2. The adjacent lateral epidermal cells, which are equivalent to the primary mother cell, become mother cells of the adjacent cells. These are divided into accessory and coronal cells, or they are functioning immediately as subsidiary cells. The lateral adjacent cells are, therefore, perigenous.

3. Also the adjacent polar cells are perigenous.

B) Syndetocheil type:

1. The primary mother cell of guard cells is, as a rule, divided into three cells the middle one of which becomes the guard mother cell. Both lateral adjacent cells are of mesogenic origin and divided, sometimes, into accessory and coronal cells. The quantity of lateral adjacent cells may be greater on one side of the guard cells than on the other one.

2. The lateral adjacent cells (subsidiary and coronal cells) of perigenous origin are missing.

3. The adjacent polar cells of mesogenous origin are missing. Polar cells of perigenous origin may often come into being.

FLORIN (1931, 1933) deserves merit for demonstrating the first time the types of history of the development of stomata. He has tried to elaborate a unitary nomenclature for stomata. It is, anyhow, a failure in FLORIN's establishments that his definition of the types of development of stomata is not unambiguous:

A) He uses the notion of primary mother cell without explaining what he means to say. By the primary mother cell he means a dermatogenous cell similar (in size, shape, plane of cell division) to the adjacent cells, of which a stoma develops. Then, in the first division, the types (A) and (B) must be contracted

as the guard mother cell is adjacent in both cases inside the primary mother cell. Is, however, the primary mother cell differring from the other dermatogenous cells, so it is difficult to distinguish between the primary mother cell and guard mother cell, and we may not speak about haplocheil and syndetocheil types.

B) FLORIN's (1931, 1933) main historical types of the development of stomata may not be generalized for more taxonomical plant groups.

C) FLORIN has not treated of the site of stoma formation, the shape and plane of cell division.

In recent times a literature treating of the histogenesis of epidermis of *Tmesopsidae* and *Pteropsidae* is unknown. The first observations were not free from some original difficulties. A lot of observations are not exact enough, they have examined a species picked out at random, without comparing families. The figures published by BRITTON and TAYLOR (1909) HILDEBRAND (1866) and PRANTL (1881) are schematic. From the few data of examination there cannot be seen wether or not the form of the development of stomata is connected with the degree of development of the family and species. On the leaf-primordium we cannot find any references in the literature concerning the site of the genesis of stomata. Any exact observations concerning the cell division and chromosomes, as well as any phographic figures, are unknown in the literature.

Materials and Methods

During my examinations (1961) I have observed the development of leaves of the following young species being in division:

1. *Psilotum nudum* (L.) GRISEB.
2. *Tmesipteris elongata* DANG.
3. *Ophioglossum crotalophoroides* WALT.
4. *Botrychium multifidum* (GMEL.) RUPR.
5. *Marattia salicina* SM.
6. *Osmunda regalis* L.
7. *Schizaea dichotoma* (L.) SM.
8. *Anemia rotundifolia* SCHRAD.
9. *Stromatopteris moniliformis* METT.
10. *Asplenium viride* HUNDS.
11. *Loxsoma cunninghamii* R. BR.

We have got the material from the hothouse and herbarium of the *V. L. Komarov Botanical Institute in Leningrad,* from the botanical gardens of the *Botanical Institute of the University in Szeged,* as well as from my own collection in Poland. The preparations were made according to the previous papers (MARÓTI 1961).

Results

A) PSILOTACEAE

1. Psilotum nudum (L.) GRISEB.

VETTER (1951) and WARDLAW (1957) have dealt with the formation of shoot and leaf of *Psilotum.* During our examinations we have observed only the development of the epidermis and stoma of the shoot. On the apical meristems of shoot the dermatogenous initials and dermatogen consist of undifferentiated,

isodiametrical cells of big nuclei. The cells are in active division. This region takes place on the apex 350—420 μ wide. The length of the dermatogenous cells is 19—24 μ. Their width is 15—22μ. The mean area of a dermatogenous cell is 378 sq. μ.

Below that region takes place the organization zone in a strife of 280—330 μ width. The guard mother cells here are formed and divided into two guard cells. The dermatogenous cell is divided into two cells of nearly equal size: the guard mother cell (Gm) and the epidermal mother cell (Em). The plane of cell division is almost straight. The genesis of stoma is euhaplocheil, its structure is acyclical.

2. Tmesipteris elongata DANG.

(Fig. 1)

Morphogenesis of the leaf

The formation of leaves of the *Tmesipteris* is treated of by BOWER (1935). Later WARDLAW (1957) has carried out, in absence of a dividing shoot, the comparison of the formation of leaves of Psilotum and Tmesipteris, on the basis of BOWER's (1935) examinations and drawings.

In the course of our examinations we have observed a shoot in vigorous division. The apex is of elliptical shape, showing several multicellular knobs. The apical meristems are well developed and somewhat defended by the lateral leaf-primordia.

We have to correct BOWER's (1935) and WARDLAW's (1957) results according to wich „the summit of the primordium in *Tmesipteris,* as seen in a radial longitudinal section, is occupied by a single larger cell of a wedge-like or prismatic form (BOWER) and unlike *Psilotum,* this apex retains its meristematic properties and potentialities".

We have, namely, found so during our examinations that the superficial cells on the multicellular knob of apex lose their meristematic potentialities after a few divisions.

The multicellular summit may have one apex (developing a foliage leaf), and it may have also three apexes if between the two apexes another apex appears rounded off and centrally as compared to the plane of cell division (developing a sporophyllum-shoot). The multicellular summit loses its divisibility at the formation of the leaf-primordium and hasn't any more role in the formation of the leaf.

A 0,5—4 mm long sporophyllum primordium functions in the same way as a shoot of limited growth. On the „apical meristems" of the sporophyllum primordium (on the multicellular knob of the apical meristems of the shoot) there develop two tubers of identical size the end of which has lost the potentiality of division and transformed into „apiculus", a spinelike outgrowth. Between the two sporophyllum knobs there appears a little centrally a rounded off sporangial knob derived from the apical meristems of the shoot. The apical meristems of the sporophyllum primordium developed in that way produces the leafstalk of sporophyllum (shoot!) and the two leaves.

Formation of the epidermis

The differentiation of shoots and leaves is reflected well by the formation of epidermis, the change of the size of epidermis cells, the place of appearance of stomata. Examining the formation of epidermis of the primordium of the Tmesipteris leaf and sporophyllum shoot we can observe a zonal arrangement. This zonal arrangement coincides in many respects with the zonal arrangement established by WARDLAW (1957) for shoots.

I have carried out many measurements and observations for determing and characterizing the single zones of the epidermis formation. I have measured the length and width of cells, their surface, the proportion of nucleus and cell content, observing also the shape of cells, the thickness of the cell-wall, the number of cells that have been dividing, the site of formation of the guard mother cells and guard cells. These examinations were carried out from the distal apex to the basis. The change of the area of cells is represented graphically from the apex to the basis. On the absciss the length of leaf is given in μ, on the ordinate the size of the cell area in the unit of $\mu^2/10$.

Taking into consideration these cytologic studies, the following zonal regions may be distinguished, in basipetal order, on the primordia of leaves and sporophylla:

(1) Distal part (apiculus of the leaf). The length of this region is 500—1400 μ. On the apical meristems of the shoot, in the time of the genesis of the multicellular knob, the superficial cells are repeatedly divided with anticlinal walls, they lose, however, soon their meristematic activity, become longer, their walls become secondarily thicker, the cells become vacuole. The size of cells decreases gradually from the summit of apiculus till the basis. The apiculus ensures the defense of cells below itself and, on the other hand, it defends the apical meristems of shoot.

(2) Sub-distal part. It takes place in a strife of 300—400 μ width below the distal region. It contains the group of embryonal initial cells. This is the centre of the meristems. The dermatogen developed here consists of undifferentiated, isodiametrical cells of big nuclei. The cells are in an intensive division. That separetes cells in the beginning also distally, later on, however, only proximally.

(3) The organogenetical part takes place in a strife of 300—500 μ width, below the subdistal region. It may be observed in that part which cells become guard mother cells and they are divided here into two, forming guard cells. The dermatogenous cells already elongate here.

(4) The sub-apical region is characterized by the vertical elongation of the epidermis cells and the powerful increase of the guard cells. The cell valls begin secondarily to become thicker and the radial walls get wavy.

These regions may, however, not be separated by cytologically rigid, sharp limits, the single regions pass over to one another.

Formation of the stomata

The guard mother cells (G. M. C.) are produced in the subdistal region with halving division, they can, however, be recognized but in the organogenetic part. In the organogenetic zone the guard mother cells enlarge, getting a little rounded off form. Then the guard mother cells are divided in two, parallelly with the longitudinal axis of the leaf, being transformed into guard cells (G. C.) The axis of division is nearly straight. We call this stoma formation to be of euhaplocheil origin.

The adjacent mother cells surrounding the guard mother cell become immediately epidermis cells. There aren't formed any subsidiary cells, therefore the stomata are acyclical.

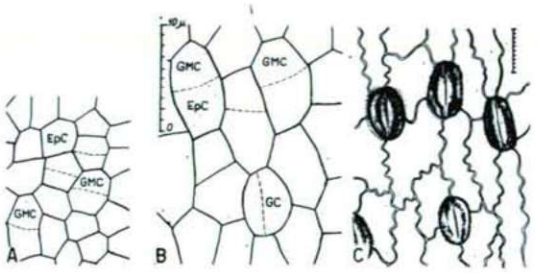

Fig. 1. Development of the *Tmesipteris elongata* DANG. stomata. G. M. C.=guard mother cell, Ep. C.=epidermal cell, G. C.=guard cell, A—B=dermatogen during division, C=consolidated epidermal detail.
Length of the consolidated guard mother cell (G. M. C.) 35—38 μ,
Width of the consolitated guard mother cell 33—38 μ,
Length of the G. M. C. during division in two: 46—57 μ,
Width of the G. M. C. during division in two: 35—45 μ.

B) OPHIOGLOSSACEAE

3. Ophioglossum crotalophoroides WALT.

The formation of the leaf shows a temporal division into sections instead of a spatial zonation. The leaf-primordium is divided in its full size. At the rim of the leaf-primordium, in a strife of 300—800 μ width, the division is more intensive than in the middle part of the leaf-primordium. At the rim of leaf-primordium the distal region is not separated, either.

The dermatogenous cell is divided with a nearly halving division into two cells: guard mother cell and epidermal mother cell. The axis of division is mildly concave or straight. The produced two cells may be of equal size or the guard mother cell is smaller than the epidermal mother cell. The guard mother cell is prevailingly produced from the distal end of the dermatogenous cell; anyhow, it may be produced also from its proximal and lateral parts. The genesis of stoma is a transition from the euhaplocheil type to the haplocheil one, its structure is acyclical. In the case of some *Ophioglossum* species accessory cells may be produced in a perigenous way.

Dimensions of the dermatogenous cells: 21x23 μ. Dimensions of the guard mother cells: 22x23 μ. Dimensions of the guard mother cell divided in two: 28x30 μ. Dimensions of the consolidated guard cell: 70x73 μ.

4. Botrychium multifidum (GMEL.) RUPR.

In the course of the formation of leaves the subdistal, organization and alongation zones cannot be separated sharply from one another. The leaf-primordium is divided, differentiated, and elongated nearly in its full size. If we examine the percentage of division from the rim of the leaf-primordium to its middle, zones may be observed which flow together.

At the rim of leaves a unilinear distal region is definitely separated. Size of the cells of the distal region: 26x35 μ. During the formation of leaf the vascular bundles are differentiated at first.

The formation of the stoma shows a temporal periodicity instead of a spatial zonation. Apart from the consolidated stomata also guard mother cells in the state of development can be found. The dermatogenous cell is divided with a nearly halving division in two cells: guard mother cell and epidermis mother cell. Between the two cells the axis of division may be straight or mildly concave. The produced two cells may be of equal size; the guard mother cell is, however, frequently smaller than the epidermis mother cell. Around the produced guard mother cells the dermatogenous cells are in an intensive division. The guard mother cell is dominatingly produced from the distal and of the dermatogenous cell; it can, however, be produced from its proximal and lateral parts, as well. The genesis of stoma is haplocheil (at some *Botrychium* species it is a transition between the euhaplocheil and haplocheil types), its structure is acyclical.

Size of the dermatogenous cell: 23x31 μ. Size of the guard mother cell: 17x23 μ. The size of the guard mother cell during division: 19x26 μ. Size of the consolidated guard cell: 38x51 μ.

C) MARATTIACEAE

5. Marattia salicina SMITH.

(Fig. 2)

The formation of the epidermis of leaves does not show any definite zonal separation. At the rim of a leaf primordium of 2—20 mm size, in the width of a cell line, the distal region may be observed. In this region 17 μ long and 13 μ wide cells may be found. The distal radial wall of the cells is thicker and the nucleus takes place in the proximal end of the cell. That layer is less divided and defends the meristematic cells below itself. Below the distal region, in 5—8 cell lines and 35 μ width, the dermatogenous initials, the subdistal region can be observed. The subdistal region cannot be sharply separated from the organization and elongation zones as even the leaf-primordium of 20 mm size is divided, differentiated, forms guard mother cells on its whole surface, and the cells elongate in the meantime, as well.

The leaf primordium produces, in the course of the division of its whole surface, dermatogenous cells in definite number, size and shape which are characteristic of the species, and in the same way guard mother cells and accessory mother cells. Then the cells elongate on the whole surface and get, with the definite size of the leaf, into the phase of ripening. In the development of the epidermis of *Marattia salicina* the temporal zonation dominates over the spatial one.

Formation of the stomata: it is preceded by production of cells of characteristic shape and formation that perform the early gas exchange. We called these cells which are unknown in literature „gas exchange hose" cells. These cells are, viewed from above, 3—5 angled (with outer tangential walls), and 4—7 small rounded off holes (perforations) of 0,5—1,5 μ diamser may be observed in their outer tangential walls. These cells enlarge towards the mesophylum, get an ovate or hose shape, their cell wall is thoroughly thin. Size of the gas-exchange hose cells viewed from above the outer tangential wall: 10x11 μ, the inner tangential wall: 26x32 μ, and their size of (radial) depth: 24—28 μ.

These cells serve as substitute for the gas exchange of the primordium that has reached a considerable size, without, however, containing developed stomata. In the gasexchange hose cells of the developed leaf quartz is stored, and so they become quartz cells.

The formation of stomata begins in the bilateral dermatogenous area of the leaf vessels, then it extends also over the intervasculary area of leaves. One of the dermatogenous cells of the organization region, in the phase of division, the primary mother cell, is divided in two cells, forming a little concave cross cell wall; the distally located cell becomes the guard mother cell, and the basally located one becomes the polar adjacent mother cell. The guard mother cell is often smaller than the polar adjacent mother cell. Around the guard mother cell the dermatogenous cells are in an intensive division. The adjacent mother cells begin to divide parallelly to the surface of the guard mother cell. The axis of division is mildly concave. The subsidiary cells are produced in a perigenous way. We call this form of the genesis of stomata to be of haplocheil type and of amphicyclical structure.

The guard mother cell is surrounded by 4—5 adjacent mother cells. From these two-three become lateral adjacent mother cells and two ones polar adjacent mother cells. Dimensions of the stoma mother cells after their formation: G. M. C. = 9x12 μ, A. M. C. = 13x17 μ.

Then the guard mother cells enlarge, the adjacent mother cells begin to divide parallelly at the periphery of G. M. C. into lateral subsidiary cell and lateral coronal cell, forming polar subsidiary cells and polar coronal cells as well.

Size of stomata in the time of the division in two of the guard mother cell: G. M. C. = 17x28 μ, L. S. C. = 5x20 μ, L. C. C. = 8x20 μ, P. S. C. = 8x26 μ, P. C. C. = 9x24 μ.

In the following the guard mother cells are divided in two by a longitudinal wall, become guard cells, then they enlarge and compress the adjacent subsidiary and coronal cells around themselves.

The adjacent protodermis cells are small, being in division, and a lot of stomata in the phase of formation may be observed around the developed stomata.

Size of the developed (mature) epidermal elements: length of epidermal cells: 40—65 μ, their width: 24—35 μ, length of the guard cells: 37—50 μ. Joint width of guard cells: 27—31 μ. Size of the gas-exchange hose cells: 13—27 μ, their number: 1. Number of stomata: 60.

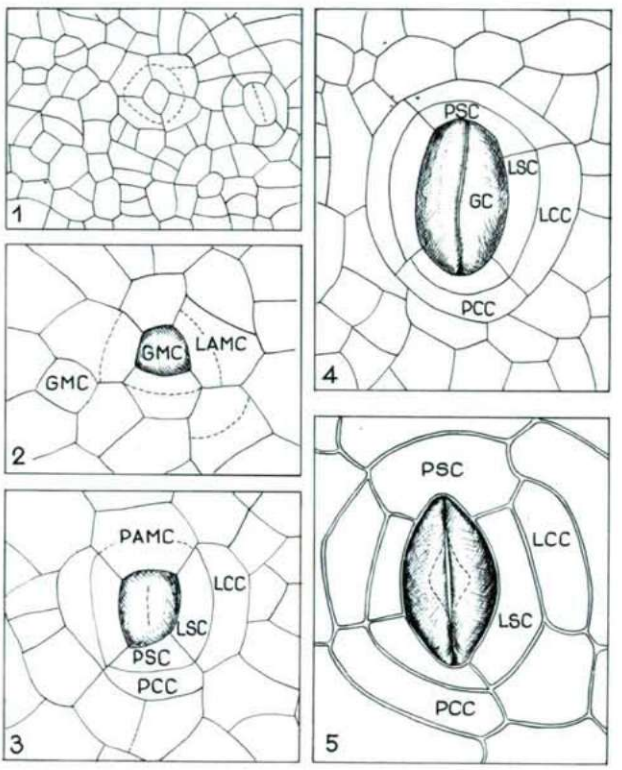

Fig. 2. Development of the stoma of *Marattia salicina*. 1—4 = dermatogenous detail in division. 5 = a fully developed stoma. G. M. C. = guard mother cell, L. A. M. C. = lateral adjacent mother cell. P. A. M. C. = polar adjacent mother cell, L. S. C. = lateral subsidiary cell, P. S. C. = polar subsidiary cell, L. C. C. = lateral coronal cell, P. C. C. = polar coronal

D) OSMUNDACEAE

6. Osmunda regalis L.

(Fig. 3, 4)

The development of the epidermis of the *Osmunda regalis* is similar to that of the *Marattia salicina*. At the rim of the leaf-primordium, in the width of 1—2 cell lines, a distal region consisting of cells of the size of 27x43 μ may be observed. Below the distal region the leaf-primordium (even that of the size of 10—15

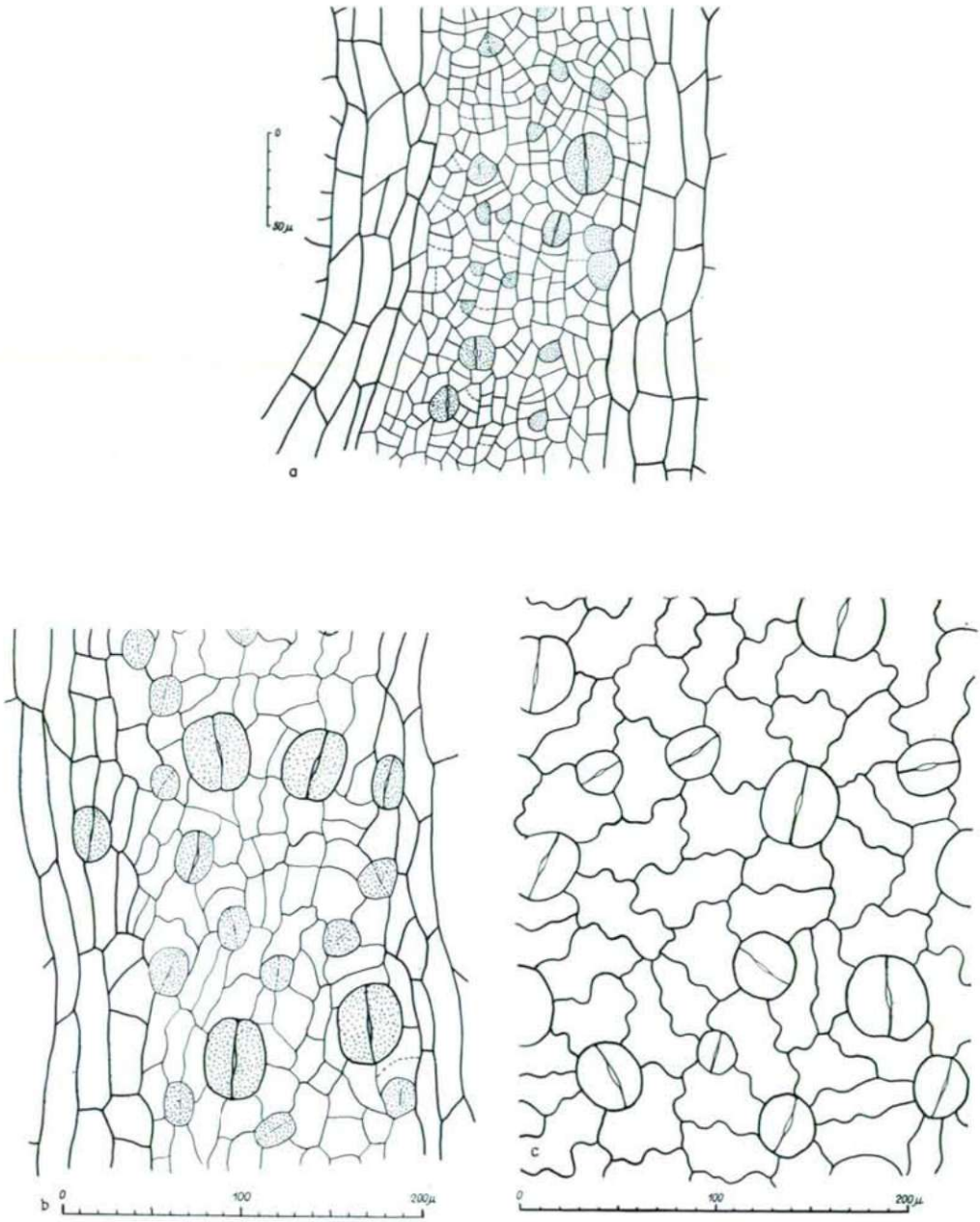

Fig. 3. Histogenesis of the epidermis of *Osmunda regalis:* a) In the intercostal field some dermatogenous cells divide in two, the distal cell becoming the guard mother cell, b) and, c) The guard mother cells enlarge and divide in two.

mm) divides on its whole surface, and protodermis cells, guard mother cells are produced in number, size and shape characterizing the species. The produced cells elongate on the whole surface, and then they get into the phase of ripening.

Formation of the stomata. The formation of the guard mother cells is similar to that of *Marattia*. From the distal end of the dermatogenous cell or from one of its corners a guard mother cell is produced by formation of concave cell walls. The basal cell of the dermatogenous cell is transformed into an epidermis cell in the way of division. The produced G. M. C. is initially surrounded by 4—5 adjacent mother cells. These cells divide at right angles to the perimeter of the guard mother cell, forming epidermis cells. Thus the developed guard cells are surrounded by 5—9 epidermis cells. If the guard mother cells are produced beside one another — what frequently occurs in the case of *Osmunda regalis* — so twin stomata are produced.

The so developed guard cells enlarge, then they divide in two by a longitudinal wall. The guard mother cell is generally smaller than the proximal epidermis mother cell. The axis of division is mildly concave. Around the guard mother cell the adjacent mother cells are in an intensive division. The adjacent mother cells frequently divide only in right angles to the developed guard. cells. There aren't produced any accessory cells. We call that form of the ontogeny of stoma haplocheil and its structure acyclical. The genesis of stoma is not purely haplocheil, it shows a transition towards the hemisyndetocheil type, as well.

Length of the guard mother cells (G. M. C.) after their genesis: 15 μ, their width: 13 μ. Length of G. M. C. before its division in two: 29 μ, its width: 23 μ. Number of stomata of a 14 mm leaf-primordium: on the summit of the leaf: 11, end the little of the leaf: 27, on the basis of the leaf: 40.

Length of the developed two guard cells: 44—63, μ, their joint width: 42—56 μ. Number of stomata: 126. Number of twin stomata: 8.

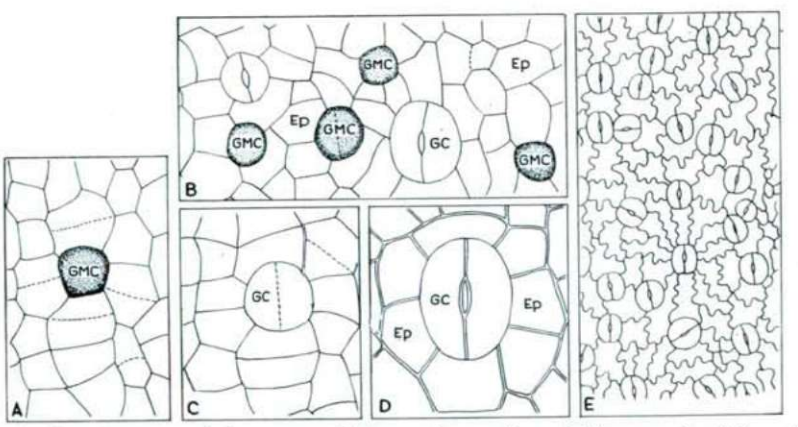

Fig. 4. Development of the stoma of *Osmunda regalis* and histogenesis of its epidermis: A = epidermis formation of the leaf-primordium of 6 mm size, B—D: that of 14 mm size, E = developed abaxial epidermis.

E) SCHIZAEACEAE

7 Schizaea dichotoma (L.) SMITH.

(Fig. 6)

In the development of the epidermis a zonal arrangement may be observed. The distal region is meristematic is nature and takes place in a cell line at the apex and two sides of the primordium. The subdistal region lies in a width of 4—5 cell lines on the apex and in 3—8 cell lines on the two sides of the leaf-primordium. This region is the centre of division. The apical part, the apical meristems ensure the longitudinal growth of leaf, producing the marginal meristems, as well as the stomatorial lath initials. The apical meristems, divide with walls forming a rigth angle to the longitudinal axis of the leaf and the marginal meristems with nearly parallel walls.

The unilinear stomatorial lath initials protruding from the surface of dermatogen, differring in shape and function and having meristematic properties are differentiated in the organization region. The guard mother cells and polar accessory mother cells develop here. The subdistal and organization regions make a transition into each other. Then, in the subspical region, the cells become much longer, getting into the phase of maturation.

Formation of the stoma: the primary mother cells of the stomatorial lath divide in two producing semicircular cell walls in the organization region. From the distal and outer tangential parts of the primary mother cell the guard mother cell and form its basal part the polar subsidiary mother cell are produced. The guard mother cells protrude from the stomatorial lath, enlarge considerably, dissolve the walls of accessory cells below themselves, and then divide in two with a longitudinal wall. The genesis of stoma is, therefore, hemisyndetocheil, its structure is diacyclical as a subsidiary cell is connected with each of the two poles of guard cells. The axis of division is thoroughly concave.

Size of the developed epidermis elements: Length of guard cells: 78—86 μ. Joint width of both guard cells: 66—74 μ. Distance between stomata: 50—124 μ. Number of stomata: 11.

8. Anemia rotundifolia SCHRAD.

(Fig. 5, 6, 7)

The formation of epidermis shows a zonal separation. The distal region takes place in a 20—35 μ wide strife on the rim of the leaf-primordium. These rim cells are 3—4 cell-lines wide, their shape in an oblong ended in a sharp point or that in a right angle to the vessels of leaves. Length of cells: 25—48 μ, their width: 8—11 μ. In the vacuolized plasma of cell there cannot be observed any chloroplastic. This region loses its meristematic property at the beginning of its formation, ensuring the defence of the subdistal region which takes place under it.

The subdistal zone is 160—200 μ wide and cannot be separated sharply from the organization region. This region is 4—6-angled, consisting of meristematic cells with big nuclei. Size of cells: 7x7,5 μ.

The organization region takes place in a 1900—2800 μ wide strife below the subdistal zone. The dermatogenous cells divide (new dermatogenous cells are produced) and are organized here. The stomata are formed here. Size of cells: 9,5x11 μ.

Below the organization region in the submarginal region the cells elongate, the chloroplastics enlarge, the nuclei are stained homogenously, the radial walls of cells have a wavy course. In the zone of maturation the cells get their definit form.

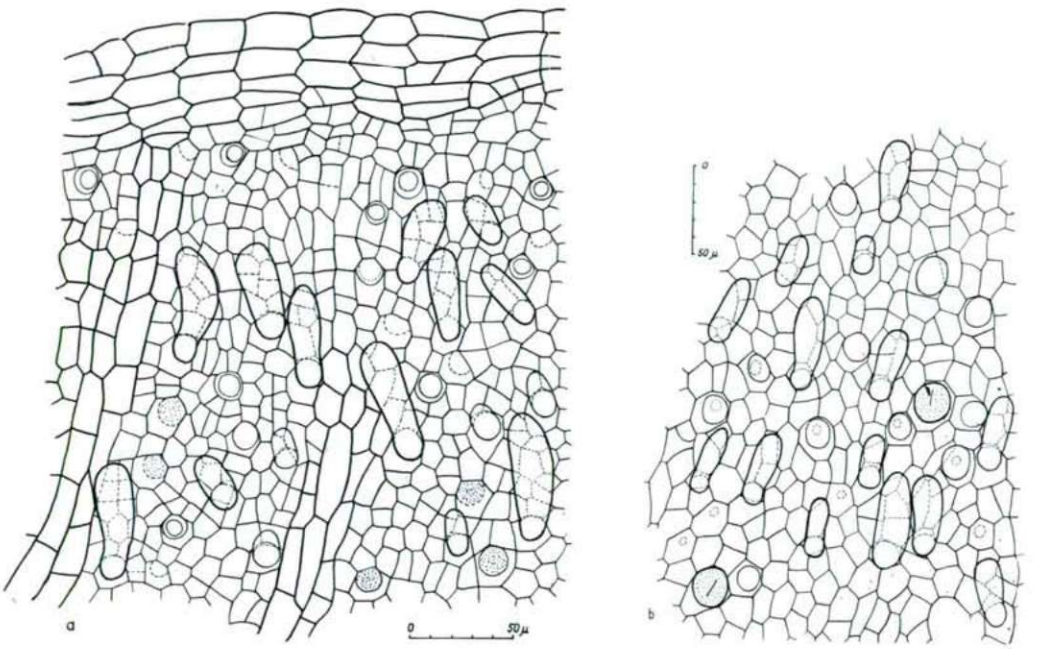

Fig. 5. Histogenesis of the epidermis of *Anemia rotundifolia:* (a) the d = distal, sd = subdistal and o = organizational regions in the lower epidermis of the leaf primordium. (b) In the organization zone the genesis of trichoma and that of the guard mother cells may be observed.

The stomata and trichomata are produced in a 1900—2000 μ wide strife. The trichomata are produced rather at the beginning of the organization region. The guard mother cell and trichoma mother cell are produced in a similar way. A spheroid knob appears centrally on the outer tangential wall of the dermatogenous cell and nearer to the distal end of the dermatogenous cell. This knob, which is at the beginning lens-like, later coniform, becomes the guard mother cell and trichoma mother cell; and the basal located cell, surrounding the G. M. C. which is at the beginning caved in and later ring-like, becomes the accessory mother cell. Later the developments of trichoma and guard cell differ from each other. The exact course of the ontogeny of the stoma can be got by the examination of

cross-sections. At some of the dermatogenous cells the outer tangential walls protuberate, the centrally located nuclei divide nearly in right angle to the surface of the leaf (in an angle of 30—45 degrees). The cell wall is formed in the shape of a hemisphere or cone towards the inside of the cell. The upper cell is the guard mother cell, the lower one is the subsidiary mother cell. The coniform G. M. C. turning with the point of cone to the mesophyllum, reaches the inner tangential wall of the subsidiary mother cell, and the cell wall dissolves at the site of touch. Therefore a round hole is produced in the subsidiary mother cell, viewed from above. Them the cells, enlarge, the guard mother cell divides in two with a longitudinal wall, the two guard cells are formed centrally, as well as a ring-formed subsidiary cell protruding from the surface and fully surrounding the former ones. We consider this genesis of stoma to be of hemisyndatocheil type and of unicyclic structure. The axis of division is thoroughly curved. It can be derived from the concave axis of division.

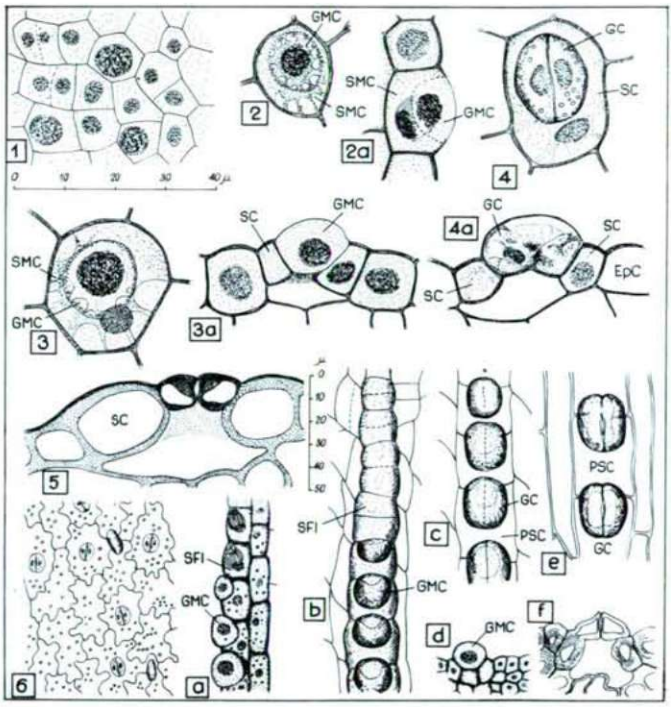

Fig. 6. The ontogeny of the stomata of *Anemia rotundifolia* (1—6) and *Schizaea dichotoma* (a—f). GMC = guard mother cell, SMC = subsidiary mother cell, SFI = stomatorial field initials, GC = guard cell, EPC = epidermal cell, ASP = air-spaces of the stoma. The degree of magnification is indicated beside the drawings. One graduation is equal to 10 μ.

The development of the trichoma differs from that of the guard cell only so far that its mother cell does not divide in two but it elongates and forms a unicellular point.

Size of the elements of the epidermis of the leafprimordium of *Anemia rotundifolia:* length of the divided GMC: 14 μ, its width: 13 μ. After division in two, the length of G is: 17 μ, the joint width of both guard cells: GC: 16 μ. Length of the mature GC: 26 μ, and its width: 22 μ. Number of guard mother cells: 20—58. Number of stomata: 18—36.

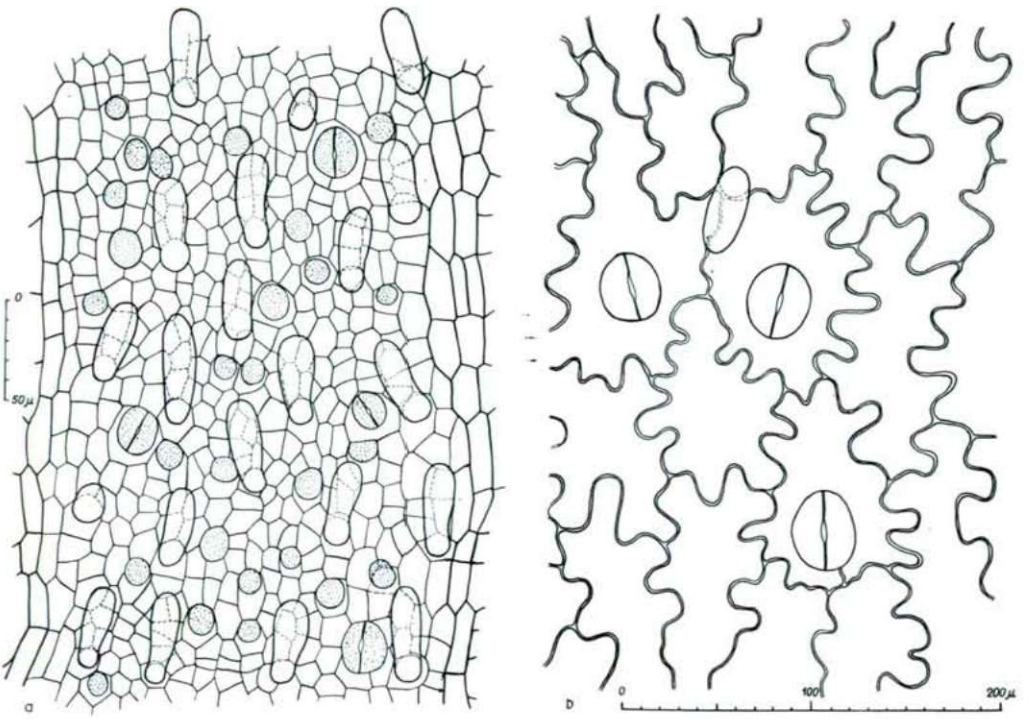

Fig. 7. Histogenesis of the epidermis of *Anemia rotundifolia:* (a) Development of the guard mother cells (dotted) in the lower epidermis. (b) The developed abaxial epidermis.

F) GLEICHENIACEAE

9. Stromatopteris moniliformis METT.

(Fig. 8)

Genesis of the leaf-slice: Two 64 μ high knobs of 8—9 cells appear in the organization region of apical meristems of the primordium of leaf-stalk, within the distance of 900—1200 μ from the apex. On the apex of the knob a big prism-shaped cell may be observed. The so produced primordium of leaf-slice elongates in the direction of apex, and later it develops in a semicircular shape by the division of the marginal meristems.

The development of the leaf-epidermis shows a zonal structure. The distal region consists of cells meristematic in nature, one cell-line wide and of 25x38 μ size. The subdistal region takes place in a semicircular strife below the rim of the leaf. In younger age, it is 200—160 μ wide, later on, after the formation of the leaf, it decreases gradually and becomes 80—55 μ wide. This region is the centre of the meristems. In the organization region (in the width of 900—200 μ) the stomata are produced in lines. The organization region decreases similarly with the growth of the leaf from the basis to the margin. In the submarginal zone the cells enlarge, the radial walls of cells become wavy. That region enlarges gradually after the formation of the leaf.

The development of stoma takes place in the organization zone. The distal end or the corner of the dermatogenous cells divides in two with a concave cell-wall formation. The distal, a little protruding cell of rounded off shape becomes the guard mother cell (GMC), the crescent shaped basal cell becomes the polar subsidiary mother cell (PSMC). The guard mother cell divides with a longitudinal cell (PSMC) derive from a common cell. The structure of stoma is diacyclic (rarely monocyclic) as two polar accessory cells join the end of guard cells. The accessory cells differ scarcely from the other epidermal cells. Size of the developed GMC: width: 20 μ, length: 16 μ. Size of the divided GMC: width: 37 μ, length: 39 μ. Size of the ripe stomata: length of the guard cells: 68—81 μ. Joint width of both guard cells: 64—71 μ. Length of the pore of stoma: 24—29 μ. Number of stomata: 73.

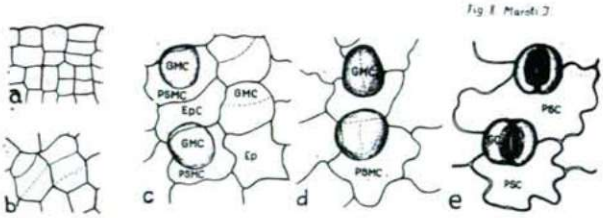

Fig. 8. Deveploment of the stomata of *Stromatopteris moniliformis*. KMC = guard mother cell, PSMC = polar subsidiary mother cell, EpC = epidermal cell.

G) ASPLENIACEAE

10. Asplenium viride HUDS.

(Fig. 9)

The development of the epidermis of the leaf demonstrates zonal structure. The distal and subdistal regions take place at the rim of the leaf-primordium, 160—70 μ wide. The two regions cannot be separated sharply, both are meristematic in nature. These regions consist of polygonal cells of big nuclei. The subdistal region is large at the beginning, later on, however, it decreases gradually while the leaf develops.

The organization region is located below the subdistal region, parallelly to the rim of leaf, in a 700—250 μ. wide strife. Here the stomata are formed and the chloroplastics appear. The organization region is large, later on, however, it decreases while the leaf develops.

The elongation and maturation zones of the epidermis demonstrate a prog-syndetocheil because the guard mother cell and the basal polar subsidiary mother wall and transforms into a guard cell. We call this ontogeny of the stoma hemi-ressive size while the leaf develops.

The formation of stoma takes place in the organization zone. The distal end or corner of some dermatogenous cells becomes guard mother cell with the formation of concave cell walls; the basal end of the dermatogenous cell remains bigger and it becomes the polar subsidiary mother cell. The guard mother cell becomes larger and divides into two guard cells with a longitudinal wall. The crescent-shaped polar subsidiary mother cell turns immediately into an accessory cell or it divides in two forming a crescent-shaped cell wall, producing so a polar subsidiary cell and a polar coronal cell. This form of the genesis of stoma is hemisyndetocheil, its structure is monocyclic with a polar subsidiary cell.

Size of GMC in the time of formation: 7,5x8 μ. Length of the dividing GMC: 15 μ, its width: 11 μ. Length of the mature guard cell: 54 μ, and the joint width of the two guard cells: 28 μ. Number of stomata: 72.

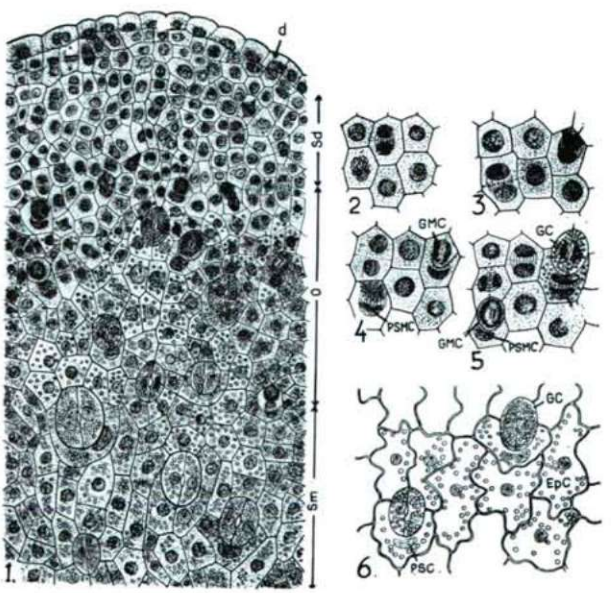

Fig. 9. Histogenesis and stoma formation of the abaxial epidermis of *Asplenium viride*. d = distal region, sd = subdistal region, o = organization region, sm = submarginal region, GMC = guard mother cell, PSMC = polar subsidiary mother cell, GC = guard cell, PSC = polar subsidiary cell, EpC = epidermal cell.

H) LOXSOMACEAE

11. Loxsoma cunninghamii R. Br.

(Fig. 10)

The formation of leaf-primordium is carried out zonally. At the rim of the leaf-primordium a less meristematic, 18—25 μ wide distal region takes place. The shape of cells is an elongated oblong in right angle to the rim of leaf. In this region the division is performed mainly with two axes of division in right angle with each other. Size of the cells: 16x22 μ.

The subdistal region is 340—60 μ wide. In the subdistal region the plane of cell division is chnging. The subdistal region is originally large, then it decreases gradually with the formation of leaf. Size of cells: 12x13 μ. This region is the centre of division.

The organization region takes place below the subdistal region in 14 000—2000 μ width. Here the mesophyllum differentiates, the stomata and chloroplastics are formed. The organizational zone cannot be separated rigidly from the subdistal zone. The plane of cell division is often concave.

The elongation or maturity zone of the leaf demonstrates a progressive size while the leaf develops.

The development of stomata takes place in the organizational zone. Some dermatogenous cells divide in two with a concave cell wall: into a lateral subsidiary mother cell ($LSMC_1$) and lateral slice mother cell (LSIMC). The latter one divides again (maybe more times) with a cell wall which is concave related to the former division wall. Thus originates the guard mother cell (GMC) of central location and the lateral subsidiary mother cell or the l. s. m. cells $LSMC_2$ or $LSMC_3$. The LSIMC is smaller than the $LSMC_1$. The $LSMC_2$ is smaller than the $LSMC_1$.

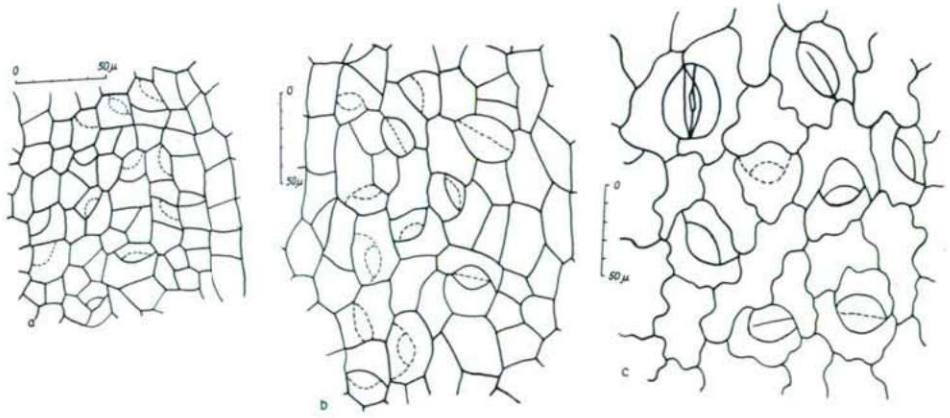

Fig. 10. Histogenesis of the abaxial epidermis of *Loxsoma cunninghamii* and the genesis of the syndetocheil stoma.

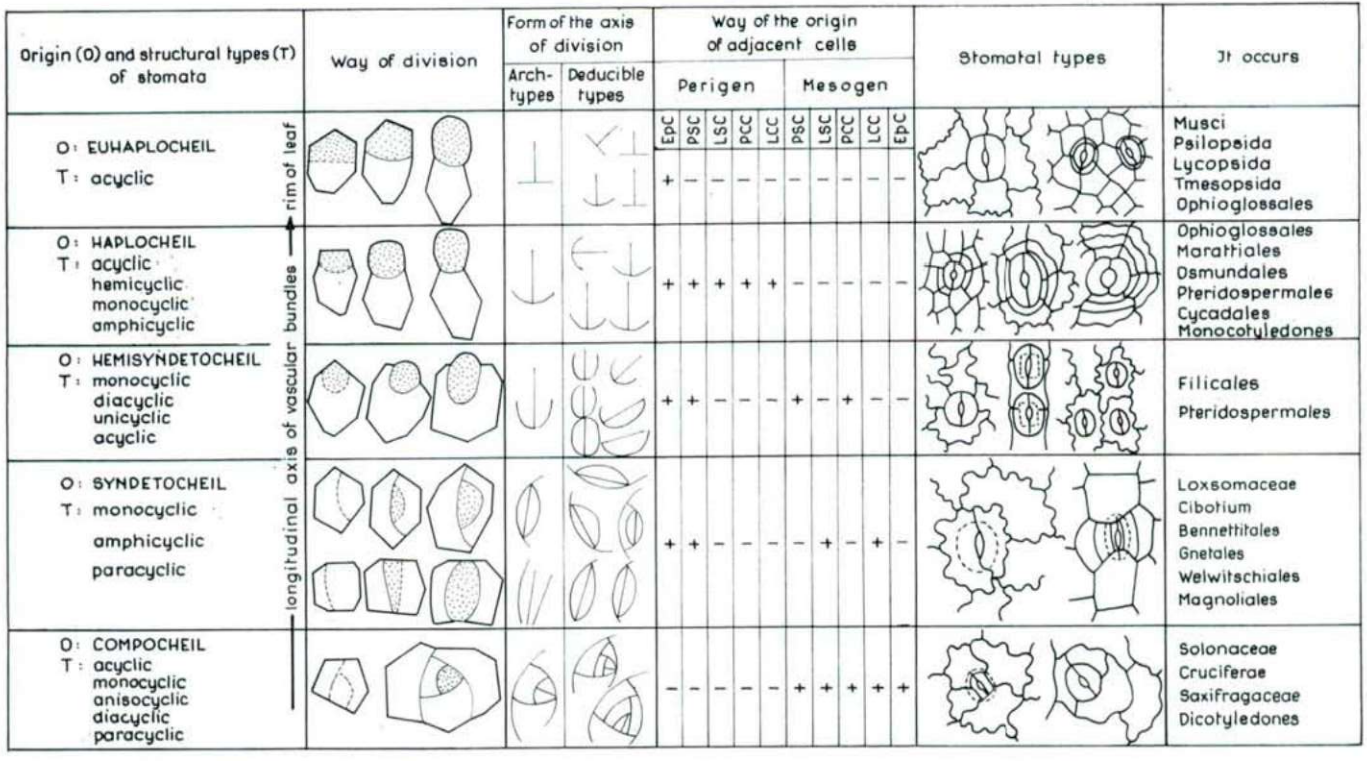

Fig. 11. Change of the area of the epidermal cells and of the number of guard mother cells (GMC) from the apex of leaf towards the basis on the leaf-primordia of *Marattia salicina*, *Osmunda regalis*, and *Anemia rotundifolia*.

The form of the axis of division is concave turned to face each other, its direction is changing, being often parallel with the longitudinal axis of the leaf vessel. From a primary mother cell dominating three, rarely four mother cells are produced.

From the lateral subsidiary mother cells lateral subsidiary cells of mesogenous origin, from the guard mother cells guard cells are produced. The guard mother cell viewed from above is similar to a biconvex lens. The Gm is gradually forced under the two lateral subsidiary cells. In the produced stoma the guard cells are thoroughly immersed.

We call this form of the stoma formation syndetocheil. The structure of stomata is paracyclic. The syndetocheil way of formation may be derived from the haplocheil one.

Size of the primary mother cell: 13x15 μ. Size of the LSMC$_1$: 8x17 μ. Size of the LSIMC: 7x14 μ. Size of the LSMC$_2$: 7x16 μ. Size of the GMC: 15x18 μ.

Discussion

A) Histogenesis of the epidermis

The development of the examined leaf of *Psilotinae* and *Filicinae* takes place in a similar way. A multicellular knot, the leaf-primordium initiative appears on the apical meristems of the shoot, and the leaf develops from that. The site of appearance, form, size, number of cells, the dominating direction of the meristematic activity of the multicellular knot are characteristic of families, species.

The development in space and time of the epidermis of the examined eleven species demonstrate a zonal structure. The regions established by WARDLAW (1957) for the structure of shoot are available also for th formtion of the leaves of *Tmesipteris* and *Filicinae*. The similar ontogenesis of both the shoots and leaves of *Psilotinae* and *Filicinae* refers to the shoot-origin of the leaves of *Psilotinae* and *Filicinae*.

Also the constitution of mesophyllum is well demonstrated by the development of epidermis. The distal regions of the leaf-primordia of the examined species are of different size, shape and meristematic activity. The distal zones of *Tmesipteris* and *Anemia rotundifolia* lose gradually their meristematic qualities after the formation of the multicellular knob.

Similarly few divisions may be observed in the distal regions of *Osmunda regalis* and *Marattia salicina*, and the division is dominating in the transversal direction. The distal regions of *Schizaea, Stromatopteris, Asplenium* have meristematic qualities. The distal region of *Tmesipteris* is large, 500—1400 μ, that of *Filicinae* is 1—4 cell-line wide.

The subdistal part is originally progressive, later on it shows a regressive size as the leaf-primordium is growing. This region is the centre of meristems. The size, form, activity, size and shape of cells of the subdistal region are characteristic of species, family. The subdistal region cannot be separated rigidly from the organization zone. In case of *Marattia* and *Osmunda*, the epidermis divides and is organized, as well, on the whole surface of leaf-primordium.

In the organizational zone the stomata, chloroplastis are produced, there begins here the elongation of cells and the nucleus gets here into equilibrium.

In the elongation and maturity zones the epidermal cells get their size and shape characteristic of the species. In the size of the dermetogenous initial cells of the subdistal region the definitive size of the epidermal cells is given. From the smaller dermatogenous cells there become smaller epidermal cells, from the bigger ones, of course, bigger cells.

B) Types of the ontogeny of stomata

Apart from the way of formation of the epidermis, the development and structure of stomata demonstrates the best how developed and differentiated the family and species are. In the course of examining the ontogeny of stomata we have taken into consideration the following factors:

(a) From how many dermatogenous cells does the stoma develop?
(b) With what kind of division is the guard mother cell formed?
(c) Plane of cell division?
(d) Where do develop the guard mother cells?
(e) From which end of the dermatogenous cell is produced the guard mother cell?
()f How do the stomata definitely develop?

On the basis of my examination we have distinguished, concerning the genesis of stomata, five main types of development:

(I) Euhaplocheil type: DC → GMC
||
EMC → EpC

(a) The dermatogenous cell divides into two cells of equal size: a guard mother cell (GMC) and epidermal mother cell (EMC) of proximal site.

(b) The plane of cell division is straight or it is nearly identic with that of the dermatogenous cells and their direction.

(c) From an epidermal mother cell only epidermal cell may develop.

(d) The cubature of the developed two guard cells is nearly the same as that of an epidermal cell.

Structure of stomata: acyclic.

Occurrence: in cases of *Psilotum, Tmesipteris, Ophioglossum, Botrychium* and *Bryophytae.*

(II) Haplocheil type: DC → GMC
EMC → EpC
perigenous PSC

(a) The dermatogenous cell divides into two cells of non-equal size: a guard mother cell and epidermal mother cell of proximal site.

(b) The plane of cell division is dominatingly in right angle to the longitudinal axis of the vascular structure of leaf, its shape being mildly concave.

(c) From an epidermal mother cell epidermal cell may develop or, with secondary division, a perigenous polar subsidiary cell (PSC).

(d) The cubature of the developed two guard cells is greater than that of an epidermal cell.

(e) Around the developed guard mother cell the perigenous division of the adjacent cell is frequent.

Structure of stomata: acyclic, hemicyclic, monocyclic, amphicyclic.

Occurrence: *Ophiolossum, Botrychium, Helminthostachys, Osmunda, Marattia, Angiopteris, Cycadales.*

(III) Hemisyndetocheil type- DC → GMC
PSMC → mesogenous PSC

(a) The dermatogenous cell divides into a smaller guard mother cell (GMC) and a bigger polar subsidiary mother cell of proximal site (PSMC).

(b) The plane of cell division is in right angle to the longitudinal axis of the vascular structure of leaf, its shape is thoroughly concave.

(c) The PSMC differs in shape, size, shape of nucleus from the other dermatogenous cells.

(d) From the PSMC a polar subsidiary cell PSC may be produced immediately (without division), in mesogenous way, or polar subsidiary cell (PSC) and polar coronal cell (PCC) in an indirect way (with division).

(e) The cubature of the developed two guard cells is generally smaller than that of an epidermal cell.

Structure of stomata: monocyclic, unicyclic, diacyclic, acyclic.

Occurrence: As a rule, at species of *Filicinae Leptosporangiatae.*

(IV) Syndetocheil type: DC → LSIMC → GMC
LSIMC → $LSMC_1$
LSIMC → $LSMC_2$ – mesogenous LSC

(a) The dermatogenous cell divides in two cells: into a lateral subsidiary mother cell ($LSMC_1$) and a lateral slice mother cell (LSIMC). The lateral slice mother cell is again dividing with a cell wall which is concave towards the former division wall, and a guard mother cell (GMC) is produced in central position plus a lateral subsidiary mother cell ($LSMC_2$).

(b) The plane of cell division is dominating parallel with the longitudinal axis of the vascular structure of leaf, its shape is concave as opposed.

(c) The $LSMC_1$ is larger than the $LSMC_2$.

(d) From the lateral subsidiary mother cell a lateral subsidiary cell (LSC) may be produced in an immediate way (without division) or a lateral subsidiary cell and a lateral coronal cell (LCC) may be produced in an indirect way (by division). There are frequently stomata at which there are on the one side LSC and

LCC, on the other one, however, only LSC. In this case the GMC is produced after the third division.

(e) The polar subsidiary cells of mesogenous origin are missing. PSC of perigenous origin may, however, be produced.

Structure of stomata: paracyclic, monocyclic.

Occurrenced: Loxsoma, Cibotium species.

$$\text{(V) Compocheiltype: DC} \rightarrow \text{LSIMC} \xrightarrow{\text{n divisions}} \begin{matrix} \nearrow \text{AMC}_1 \\ \text{GMC} \\ \searrow \text{AMCn} \end{matrix}$$

(a) The dermatogenous cell divides into two cells: into adjacent mother cell one (AMC_1) and a slice mother cell (LSIMC). The slice mother cell separates after n divisions into a guard mother cell (GMC) and n adjacent mother cells (AMCn).

(b) The plane of cell division is changing, it is generally in right angle to the axis of division, its shape is mildly concave, being turned back.

(c) From the adjacent mother cell there may be produced an epidermal cell, subsidiary cell, and coronal cell in a direct or indirect way.

(d) The adjacent cells are of mesogenous origin.

Structure of stomata: acyclic, anisocyclic, monocyclic, diacyclic.

Occurrence: *Solanaceae, Cruciferae, Saxifragaceae.*

From the *Angiosperms* we have observed the ontogeny of the stoma of these few families. We suppose that the majority of the dicotyledonous plants belong into this type from the point of view of the ontogeny of stomata.

Fig. 11. Types of the history of development of the genesis of stomata.

Summary

(1) The leaf formation of the examined eleven species demonstrates a zonal structure in space and time. The regions ascertained by WARDLAW (1957) for the organization of shoot are valid to the formation of *Tmesopsida* and *Pteropsida* leaves, as well. In the course of the organization of the leaf primordium, distal, subdistal, organizational, lengthening, and ripening zones were distinguished.

(2) The histogenesis and structure of leaf mirrors he degree of he development of species, as well its natural taxonomical place.

(3) The skin tissue is the most suitable of the tissues of leaves for comparative histogenetic examinations, especially concerning the development and structure of stomata.

(4) In the course of our examinations, five archetypes of the history of development of the genesis of stomata have been distinguished: euhaplocheil, haplocheil, hemisyndetocheil, syndetocheil, and compocheil ones.

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EFFECT OF WAVELENGTH COMPOSITION OF LIGHT ON THE ACCUMULATION OF NITROGENOUS COMPOUNDS

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(Received February 15, 1967)

Introduction

Experiments reporting on the effect of different ranges of wavelengths have pointed out, more or less unanimously, that shorter wavelengths of light tend to promote the synthesis of nitrogenous compounds. To account for these observations several authors suggest that path of the photosynthetically assimilated carbon is influenced by variations in wavelength (CAYLE—EMERSON, 1957; NICHIPOROVICH, 1958; VOSKRESENSKAYA—GRISHINA, 195,; HAUSCHILD et al., 1962 a, b; TOLBERT, 1963; VOSKRESENSKAYA, 1965).

At an explanation like that presents no small problem a fact that treatments of different wavelength, even if applied with an identical light intensity or incident quantum number, result in a different CO_2 fixation, resp. accumulation of dry matter. The quantitative changes mentioned above goes, in itself, with the change of the products of photosynthesis (NICHIPOROVICH, 1963; VOSKRESENSKAYA—GRISHINA, 1958, 1959; HAUSCHILD et al., 1962a).

Exeperiments ensuring an identical photosynthetic intensity did not give any unambiguous answer. Thus HAUSCHILD et al. (1962a) mention the decrease of glycin, serin and glycolic acid simultaneously with the increase of asparaginic acid under the influence of blue light, which changes have compensated one another. TREGUNNA et al. (1962) have observed a marked decrease of glycin and, in a smaller degree, that of serin in blue light. VOSKRESENSKAYA and GRISHINA (1959) have obtained, even under such conditions, in the leaves of beans, and VOSKRESENSKAYA and NECHAEVA (1966) in those of barley a higher protein content in blue light.

In the experiments to be reported now we have investigated the effect of different wavelength composition of light on the accumulation of nitrogenous compounds.

Material and methods

Bean plants (cult. Surecrop) were grown under fluorescent lamps of different wavelength distribution of radiant energy till an age of five weeks. Illumination has varied between the limits 30 000—120 000 erg/sq. cm. sec. In our equipment (HORVÁTH, 1964) the temperature

has changed between 15—25 °C, the vapour content between 40—70 percent, in a given daily rhythm. The plants were grown in sand culture using PRYANISHNIKOV's nutrient solution.

From the dry material of plants water-soluble and protein nitrogen fractions, precipitated by TCA were isolated. After digestion by sulfuric acid nitrogen determination have been carried out by direct Nesslerization (KELLEY et al., 1946).

Results

The soluble nitrogen content, taking place by an identical incident quantum number, is illustrated in Fig. 1. Although the values in several organs are different and depending also upon light intensity, the amount of the soluble nitrogen fraction has decreased in every organs in the order of green, red and blue light. Thus wavelength composition has produced unequivocal effect.

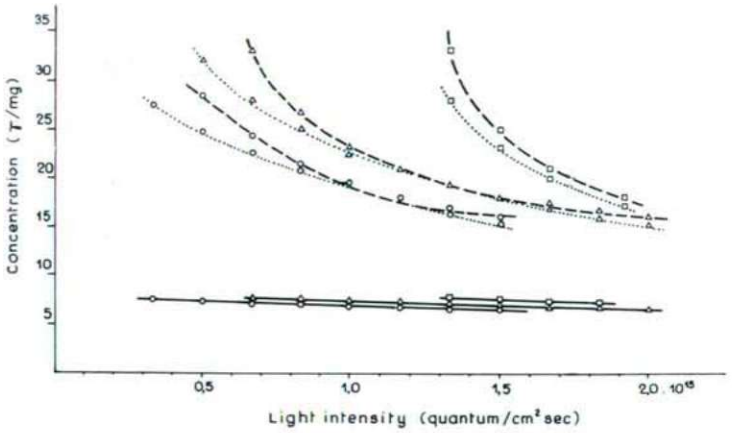

Fig. 1. Effect of wavelength composition of light on the soluble nitrogen content. o, blue; △, red; □, green light; ——, root; - - -, stem; . , . , leaf.

The protein nitrogen content has not been influenced neither by wavelength composition nor by light intensity. Only the difference of several organs was measured. The protein nitrogen level has varied about 14 γ/mg in the root, 9 γ/mg in the stem, and 20 γ/mg in the leaf.

Discussion

As the protein nitrogen has not shown any change the soluble nitrogen fraction will be considered by interpreting the effect of wavelength composition of light.

Under our experimental conditions it was observed, in the case of an identical accumulation of dry matter, that soluble nitrogen content is not touched by variations in wavelength (Fig. 2). The data also indicate that increasing dry matter accumulation accompanies the decrease of soluble nitrogen content.

Accordingly we consider the effect of wavelength composition in the fact of changing the course of the accumulation of dry matter (Fig. 3). If the increase in dry matter production has an effect on the proportion of some compounds, so the influence of wavelength composition is manifest. In the present case this is true of the soluble nitrogen fraction. If the increase in the amount of produced dry matter does not show any relationship to the proportion of

1) As related to an identical incident quantum number, the soluble nitrogen content has decreased in every organ in the order of green, red and blue light.

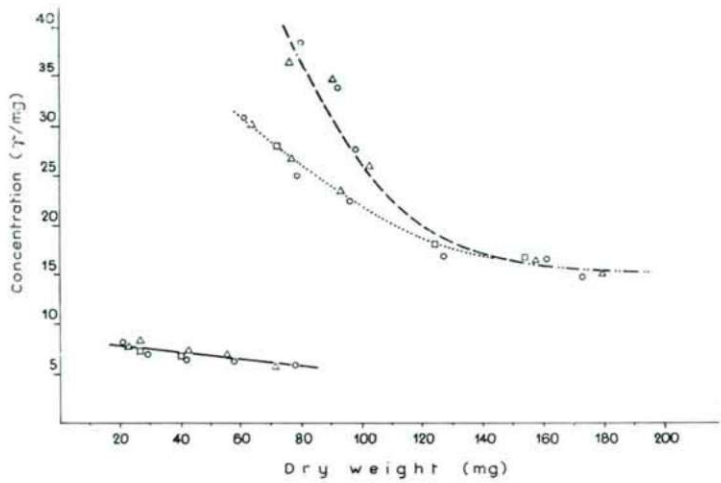

Fig. 2. Effect of dry matter accumulation on the soluble nitrogen content. Marking is the same as at Fig. 1.

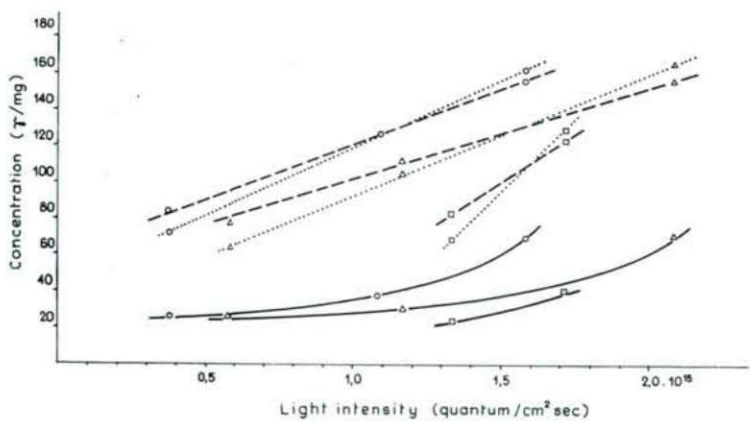

Fig. 3. Effect of wavelength composition of light on the accumulation of dry matter. Marking is the same as at Fig. 1.

2) The level of protein ntirogen remained constant.

3) We interpret the effect of wavelength composition of light in the fact of changing the course of the accumulation of dry matter.

any compound, so wavelength composition has no effect. That has been experienced of the protein nitrogen content.

The high ntirogen content, produced in blue light and cited in the literature, could not been found in our experiment. We explain this fact so, referring to data in Fig. 2, that dry matter production in blue light was greater than in red and green ones. These data are in accord with the observations of HOOVER (1937), KLESCHNIN (1960) and MCLEOD (1961). VOSKRESENSKAYA (1952, 1961), on the other hand, has found consistently a higher photosynthetic activity in red light.

Summary

Investigatigations on the effect of wavelength composition of light upon the accumulation of nitrogenous compounds have been carried out growing plants under controlled conditions for several weeks. The investigations indicate:

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GEOTRICHUM NOVAKII N. SP.

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Several yeasts were isolated from decaying fruits in Egypt. The strains were identified as *Torulopsis westerdijkii* VITÉZ et NOVÁK, *Torulopsis sake* (SAITO et OTA) LODDER et KREGER van RIJ, *Paratorulopsis ingeniosa* (DI MENNA) NOVÁK et ZSOLT, *Paratorulopsis maris* (VAN UDEN et ZO BELL) NOVÁK et ZSOLT (names according to the system of NOVÁK and ZSOLT, 1961), *Geotrichum candidium* LINK and a *Geotrichum* strain different from the *Geotrichum* species described till now. This strain is described in the following.

Material and methods

The strain was isolated from peach *(Prunus persica)* and is maintained in the collection of the National Research Centre, Cairo.

According to the previously expressed opinion of one of the authors (ZSOLT et al., 1961; NOVÁK and ZSOLT, 1961), *Geotrichum* may be considered as a yeast-like fungus, therefore the methods proposed by the Dutch School of yeast taxonomy (LODDER and KREGER VAN RIJ, 1952) were applied for characterizing the new strain.

Results

Growth on malt agar: After 3 days, at 25 °C, long dichotomically branched, septate, 3—4 μ thick hyphae and 3—4 x 2—3 μ more or less rectangular arthrospores (Fig. 1).

After 1 month, at 25 °C, the streak culture was white, dull, flat.

Slide culture on potato agar: similar microscopis picture as on malt agar.

Sporulation: No ascospores were formed on Na-acetate agar.

Fermentation: No fermentation was observed.

Assimilation of sugars: Glucose, galactose and maltose were assimilated. Assimilation tests with sucrose, lactose and raffinose gave negative results.

Assimilation of potassium nitrate: negative.

Growth on ethanol as sole source of carbon: positive.

„Starch" formation: absent.

Carotenoid pigments: not produced.

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Discussion

Macroscopic appearence of the colonies, mycelial growth and production of arthrospores, lack of budding and of ascospores and absence of fermentation show that our strain belongs to the genus *Geotrichum* LINK.

Yeasts showing different characteristics during the standard identification process are generally considered as members of different species. Although this assumption is probably not entirely correct, in the present stage of the taxonomy of fungi it seems useful to be accepted. In the special case of our new strain this is all the more reasonable because this strain differs from the other *Geotrichum* species in the assimilation pattern of sugars which characteristic is a rather constant one (NOVÁK and ZSOLT, 1964). Therefore we describe the strain as a member of a new species: *Geotrichum novakii*. The specific name „novakii" refers to DOCTOR E. K. NOVÁK a distinguished scientist of yeast taxonomy.

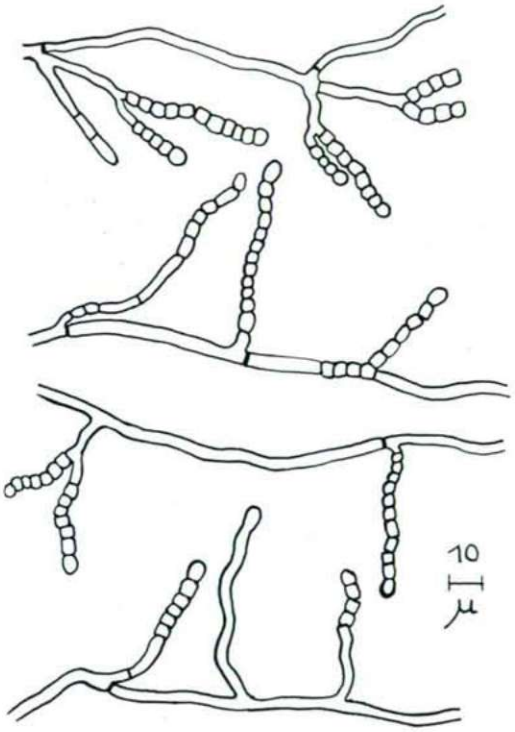

Figure 1. *Geotrichum novakii n. sp.* Growth on malt agar

MORENZ (1964) distinguishes further *Geotrichum* species on the basis of morphological characteristics and assimilation tests with further carbon sources. As regards this method, there cannot be raised any logical objection. Nevertheless, we maintain the methods of the Dutch School generally used by yeast taxo-

nomists in distinguishing species although we are conscious that, as a result of a rather wide-spread mutual agreement of scientists, also these methods are more or less provisional.

Geotrichum gracile (WEIGMANN et WOLFF) Windisch accepted by MORENZ seems to be closest to our strain but *Geotrichum gracile* has only doubtful galactose and maltose assimilation and has no dichotomically branched hyphae.

A comparison of the most important characteristics of the new species and of the other *Geotrichum* species is given in Table I.

Table I. Comparison of the most important characteristics of the *Geotrichum* species

	assimilation of							
name	glucose	galactose	sucrose	maltose	lactose	raffinose	ethanol	potassium nitrate
G. linkii VÖRÖS—FELKAI	+	−	−	−	−	−	+	−
G. candidum LINK	+	+	−	−	−	−	+	−
G. novakii n. sp.	+	+	−	+	−	−	+	−
G. matalense CASTELLANI	+	+	+	+	+	+	+	−

Caretta (1963) considers *Geotrichum linkii* as a synonym of *Torulopsis zeylanoides*. This must be a mistake because *Geotrichum linkii* shows no budding but has all the morphological characteristics of a *Geotrichum*.

Little can be said about the phylogenetical position of the *Geotrichum* species. A possibility of their deduction from the perfect genus *Endomyces* is, anyhow worth of consideration.

Diagnosis

Geotrichum novakii n. sp.

In agaro maltato septatae hyphae (3—4 μ) et arthrosporae (3—4 x 2—3 μ). Cultura (post unum mensem, 25 °C) albida, parum nitens, plana.

Pseudomycelium nullum. Sporulatio nulla. Fermentatio nulla.

Glucosum, galactosum, maltosum et alcoholum assimilantur, saccharosum, lactosum, raffinosum et nitras kalicus non assimilantur.

„Amylum" et pigmenta carotenoidica non synthetisantur.

Isolata ex fructo *Pruni persici*.

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A NEW INDICATOR OF ABNORMAL AMINO ACID AND PROTEIN METABOLISM IN DISEASED PLANTS: RICE, POTATO, SOYBEAN, TOBACCO

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(Received September 26, 1966)

Introduction

The free amino acid content of plant organisms may be increased by means of a large N or P supply, or in consequence of certain diseases (BIDWELL et al., 1964; FARKAS and KIRÁLY, 1961). By paper chromatography we have looked for a physiological indication to demonstrate beyond any doubt if or not the increase of the free amino acid content is a consequence of some infection.

An unknown substance, lying at 0,64 Rf between the spot of γ-amino butyric acid and valine, was detected first in the chromatogram of rice (PÁLFI, 1964). Fixed with copper nitrate, this substance, in distinction to amino acids, gives a blue colour instead of a red one. Therefore, the unknown substance has been designated by us as „blue subtance" and its indication as the „blue test." In the case of rice, it appeared that under conditions promoting the occurence of diseases (PÁLFI, 1956) or even provoking them artificially (PODHRADSZKY, 1961) the more resistant foreign varieties contained less of the „blue substance" than the less resistant Hungarian types. Therefore, the test may be suggested as a physiological indication to select disease-resistant varieties.

By preparing an amino acid-free protein extract of diseased leaves and hydrolizing it with hydrochloric acid, the „blue substance" cannot be found among the amino acids produced. Thus it is not a component of protein. When the free amino acid extracts containig the „blue substance" are hydrolized, all the amides and peptides in them decompose but the „blue substance" remains. Thus it is neither an amide nor a peptide. Since the „blue substance" has been detected in wheat, as well (PÁLFI, 1965), our investigations were extended to other plants, too.

Materials and methods

Ascending one-dimensional papers (WHATMAN No 1) have been slowly developed by being cooled in a 2:1:1 solvent of butanol-acetic acid-water. In the second dimension a phenol-water mixture (4:1) has been applied. Final development has been secured by ninhydrine at 98 °C. The alcoholic extract of 1 g fresh substance or 200 mg substance dried at 65 °C served as starting material. Leaves of healthy and diseased plants were collected from the

same parcels during flowering. 12 Hungarian and foreign rice varieties were studied among others the *Hungarian Dunghan shali*. Potato was from the sort of *„Tompa rózsa"*; soy-bean *„Iregi korona"* and tobacco *„Szabolcsi"*. The diseases were: *„blast disease"* at rice *(Piricularia oryzae* CAV.), *„leaf roll virus"* (*Corium solani* HOLMES) at potato; *„soybean mosaic"* at soy-bean and *„Marmor tabaci var. vulgare Holmes"* at tobacco.

Results

Figure 1 shows that the „blue substance" is present besides rice in the leaf-extracts of diseased potato and tobacco, as well. At the same time, the extracts of healthy plants collected from the same parcels as the diseased ones did not contain the „blue substance". Figure 2 illustrates that the blue spot can be found in the chromatogram of the leaf extracts of virus-infected soy-bean, too. And the „blue substance" couldn't be found in the case of the healthy control, either. It can be seen that „blue sbustance" reaches the largest Rf value in the second dimension, i. e., in phenol-water solvent.

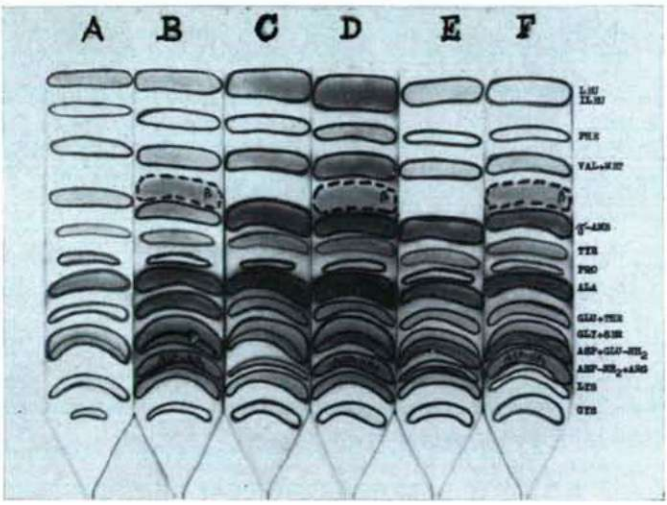

Fig. 1. Chromatogram of leaf extracts of healthy and diseased plants. A = healthy rice; B = Piricularia infected rice; C = healthy potato; D = virus infected potato; E = healthy tobacco; F = virus infected tobacco. β = blue spot, unknown.

Figure 3 shows the two-dimensional development of the leaf extracts of virus-infected tobacco — the „blue substance" appears here, too. In this figure as well as in the previous ones it is obvious that the „blue substance" appearing, also asparagine can be found in a considerable amount. Further it was experienced that i diseased plants where γ-amino butyric acid was present in a larger amount, more „blue subtance" could be detected.

It may be seen in figures 4 and 5 that the „blue substance" appears only in the thin layer chromatogram of the diseased potato.

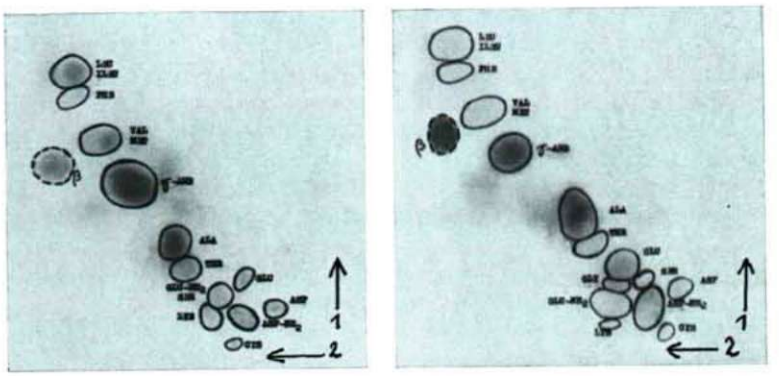

Fig. 2. Chromatogram of extract from virus-infected soybean leaf. β = the „blue spot".
Fig. 3. Chromatogram of extract from virus-infected tobacco leaf. β = the „blue spot".

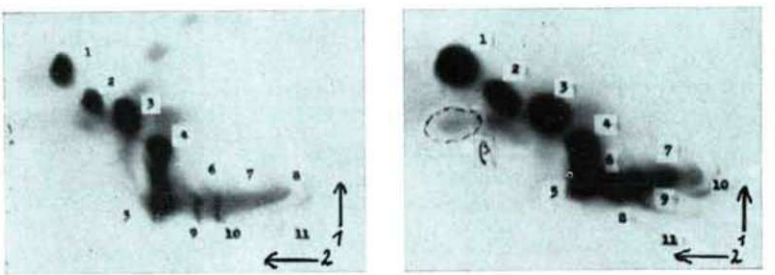

Fig. 4. Thin layer chromatogram of healthy potato. 1 = Leu + Ileu; 2= Val + Met; 3 = — Amb; 4 = Ala + Thr; 5 = Glu — NH_2 + Arg; 6 = Glu; 7 = Ser + Gly; 8 = Asp; 9 = Lys; 10 = Asp — NH_2; 11 = Cys.

Fig. 5. Thin layer chromatogram of diseased potato. 1—7. numbers as in Fig. 4; 8 = Lys; 9 = Asp. — NH_2; 10 = Asp; 11 = Cys; ß = blue spot.

From the leaves of rice containing „blue substance" a tapwater extract has also been prepared and inoculated with a pure culture. Bacterii of seven species, among them *Bac. subtilisATCC 6633* and *Escherichia coli 0 111* consumed the amino acids at 30 C° in seven days but the „blue substance" remained unchanged. It follows from this that this substance is no 1-amino acid and is not toxic.

It was detected that the largest amount of the „blue substance" could be found in the leaves of plants, the stem containing less and the roots and seed the least of it. In the lower leaf storeys of diseased plants there was less „blue substance" than in the younger upper leaves. Thus the „blue substance" appeared in a large amount where the protein metabolism was more intense. In the case of rice, during ripening of the corn when the protein synthesis was markedly decreassed in the still green but already old upper leaves, too, the „blue substance" could hardly be pointed out from the extracts. These data indicate that the appearance of the „blue substance" is connected with an increased protein metabolism.

The blue spot can be detected not only in plants mentioned in this paper but on the leaves of other diseased plants, as well. Thus recently it has been detected also in the chromatograms of extracts of virus-infected bean, sunflower and virus and *Phytophtora* infected *Solanum laciniatum* AIT. medicinal herb. These plants, however, have not been studied in detail.

Summary

In the course of the study of the free amino acid spectrum of diseased rice, soybean, potato and tobacco leaves an unknown compound has bean detected, the appearance of which is correlated with an abnormal physiological state of the plants, or with some disturbances in the amino acid — protein metabolism.

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CHANGES IN THE AMINO ACID AND PROTEIN METABOLISM OF RICE AS A RESULT OF DISEASES

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Introduction

Hungary is the northest area in Europe where rice can be grown. There are years when, owing to the prolonged cool weather and the lack of sunshine in summer, the rice diseases are rather frequent. In such years even a 40—75 p. c. lowering of productivity was observed (e. g., in 1949, 1954. 1955). After 1950 the rice growing has taken more and more areas. For a long time there had prevailed a wrong view alleging that rice grows well also in our alkali soils. Our experimental results, obtained so far, added, however, a lot to make it clear that our Hungarian-bred rice sorts were not salt-resistant and were unsuitable for alkali soils (Pálfi 1965 b). At rice growing in alkali soils, in case of unfavourable summer weather, the occurrence of, and the damage caused by, diseases is much more expressed than among plants growing in other solis. Just the widespread appearance of rice-diseases makes possibile to study the damaging factors.

In leaves of diseased rice plants and of plants overfed with N, a compound of unknown composition was demonstrated by paper chromatography (PÁLFI 1964 a, 1965 a).

After the reaction with ninhydrine and fixation by copper salt, this substance gives a blue colour instead of the red characteristic for the amino acids. In further investigations it appeared that the size and intensity of the blue spot increased parallel to the extent of unilateral N-nutriton before sowing (PÁLFI 1964 b, c).

The appearance of the blue spot is connected with disturbances in the metabolism (probably in the amino acid and protein metabolism) since the spot could always be detected in diseased rice, namely in an amount proportional to the extent of disease (PÁLFI 1965 a). During studies of healthy rice grown in the fields throughout the former years, this spot has not been demonstrated in any case (PÁLFI 1963).

The present paper set itself the task of ascertaining whether or not the blue spot, named by us „blue substance" and designed by „β", can be demonstrated in other parts of the rice plant apart from the leaves. Our previous results had been obtained on a single rice sort, the Hungarian-bred Dunghan shali. In the course of the present experiments other Hungarian and Soviet rice sorts have been investigated as well. If the appearence of the blue spot is connected with similar con-

ditions in other rice sorts, as well, its demonstration, i. e., the „blue-test" has a general validity.

In the course of our previous experiments it had already been demonstrated that the amino acid (especially the glutamine and asparagine) content in the rice leaves may increase not only as a result of an abundant N-nutrition but this increase may appear also as a consequence of any influences disturbing the protein synthesis (PÁLFI 1965 b). Similar results have also been published by other authors (PLESKOV 1957, FARKAS 1963, BIDWELL et al. 196, FELDMANN and HANKS 1964, SEHGAL and BOONE 1964).

Material and methods

The experiments were carried out by the researchers of the State Institute for Qualification of Sorts and Agricultural Technology (Országos Mezőgazdasági Fajta- és Termeléstechnikai Minősítő Intézet; OMFTMI). Physiological experiments were performed in the State Plantation, Kopáncs. The purpose of experiments made by OMFTMI was to provoke rice diseases by addition enormous amount of nitrogen before sowing, by excluding the light artificially and through infections (VÁMOS 1959, ZSOLDOS 1962). This method is suitable to select brusone-resistant sorts (PODHRADSZKY 1961).

Dunghan shali was grown without fertilizer (control) and having 3 and 6 q ammonium sulphate per cadastrial acre, respectively (1 cadastrial acre = 0, 5755 ha). Moreover, samples were taken from the following sorts: *„Pannoryza 2"*, *„Bánát 725"*, *„ H6"*, *„H 9"*, *„Káka 203"*, *„Szarvas 63"* Hungarian-bred rices, and *„Cuban 3"*, *„Krasnoarmenskij 312"*, *„Dubovskij 129"*, *„Krasnodarskij 424"*, *„Uzoros 17"* Soviet sorts. Samples were taken six times: during shooting, blooming, embryogenesis and further three times during ripening. Samples were also taken in the previous year from Dunghan shali in the State Plantation, Pale.

As published by us (PÁLFI *1964 b*) and others as well. (MOTHES *1960*, YOSHIDA *1961*, COIC et al. 1963, BOODSON et al. 1964, LIZANDR and BROVCUNA 1964), the nutrient (NPK) content of leaves differs in shoot by leaf storeys. Therefore, the different leaf storeys were separatly analyzed, and the results obtained on the upper second leaves compared with. Most of the seedlings had only three living leaves.

In each case fresh leaves fixed and dried at 65 C° were analysed. The ascending paper-chromatographic analysis of the 50 p. c. ethanol extracts was carried out on Sch.-Sch. 2041 b and WHATMAN No 1. paper. Butanol-glacial acetic acid — water solvent has been applied in a ratio of 2 to 1 to 1. The running was slowered by cooling. (2 C°) and sometimes the running was repeated several times. In two-dimensional development phenol-water (4 to 1) served as a second solvent. The demonstration of the compounds was performed with ninhydrine, isatine, alioxan and folin reagents. In the identification of spots the method of the universal standard mixture, elaborated by us, had been applied (see Figs. 1 and 2;) (PÁLFI 1964 b, c and 1965 a). For the quantitative determination the spots were fixed by copper-salt, eluted and then subjected to photometry (SZALAI 1957). The extinction value of the eluted standard series yielded the calibration curve. Hydrolysis has been performed with 6 n hydrochloric acid for 24 hours at 105 C°, the samples being sealed in glass tubes.

Results

The Hungarian-bred Dunghan shali, as it is well known, is not resistant to diseases. The weather was unfavourable enough for the occurrence of different diseases. There were quite a number of rainy, cool days and the clear days were followed by a strong cooling at nights. Sunshine hardly intruded the plantstand, which was very dense owing to the great N-content. The flooding water was very cold (15—17 C°) even in the sunny hours at noon. Leaves of rice supplied with great amount of N before sowing have been longer and wider than those of the controll. Their colour was dark green, their tissue loose, fragile. During clustering

there appeared lodged, diseased spots in the rice fields. The leaves of such shoots, as in the former years (PÁLFI 1964 b, c), showed well the brownish-grey spots of brusone *(„Piriculariosis")*. Diseased shoots and smaller spots of diseased plants appeared as well in the rice fertilized with 3 q N per cad. acre.

In Fig. 1 three samples of Dunghan shali, supplied with different amounts of N are to be seen. The blue spot framed with broken line („β") on the chromatograms is situated between valine and γ-aminobutyric acid. As it can be seen, the blue spot appeared only in rice supplied with 3 and 6 q N per cad. acre ammonium sulphate but not in the control. In Fig. 1. the formerly described universal standard mixture (PÁLFI 1964 b, c, 1965 a) can be found in three concentrations and it is possible from them to make quantitative estimation of the investigated extracts.

Starting from identical amounts of the different parts of rice and using the same amount of extracts for chromatography, it was impossible to compare with one another the results due to the very different intensity of the spots. Therefore a simple amount of the leaf- and cluster-extracts, twofold amount of the stem-extracts and a fourfold amount of the root-extracts were carried on the paper. In Fig. 2 it can .very well be seen that the blue substance appears in a larger amount in the leaf and in the stem, less in the root and the least in the cluster. Furthermore it is striking to see in this cromatogram the large asparagine content of the cluster which may refer to a strong protein synthesis.

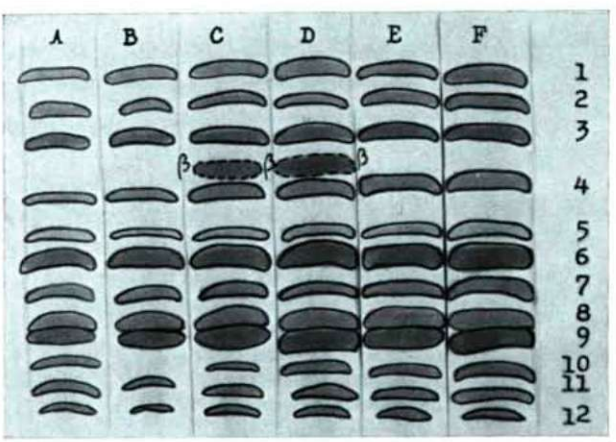

Fig.1. Chromatogram of leaf-extracts of Dunghan shali with different N-supply. Blooming. „A" : universal standard with 25 μg total amino acid content. „B" : rice without fertilizer. „C": 3 q ammonium sulphate per cad. acre. „D": 6 q ammonium sulphate fertilizer par cad. acre. „E" and „I": universal standard mixtures with 50 and 75 μg total amino acid content. „β": blue spot, the unknown substance.
Composition of the universal standard in the case of 50 μg total amino acid:

1. Leu	1,5 μg	7. Glu+Ser	6,0 μg
2. Phe	3,0 „	8. Gly+Glu—NH_2	3,5 „
3. Val	1,5 „	9. Asp	10,0 „
4. γ-Amb.	1,5 „	10. Asp—NH_2	8,0 „
5. Pro	5,0 „	11. Lys	1,0 „
6. Ala	5,0 „	12. Cys	4,0 „

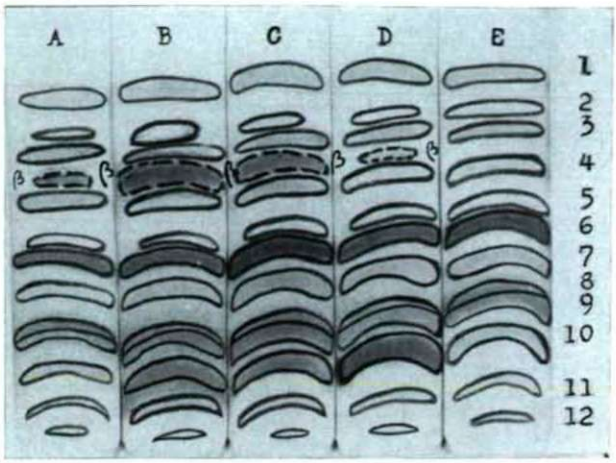

Fig. 2. Chromatogram of extracts of different parts of the rice plant in case of a great amount of N-supply. Dunghan shali in blooming. „A": root. „B": stem. „C": leaf. „D": cluster. „E": universal standard with 50 μg total amino acid content. „ß": blue spot. Numbers as in Fig. 1.,

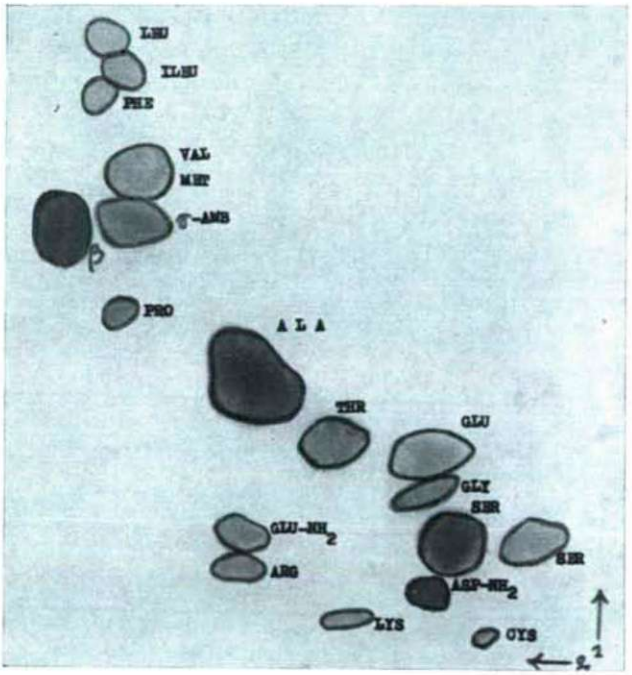

Fig. 3. Chromatogram of the leaf extract of rice with abundant N-supply. Dunghan shali, blooming. „ß": blue spot.

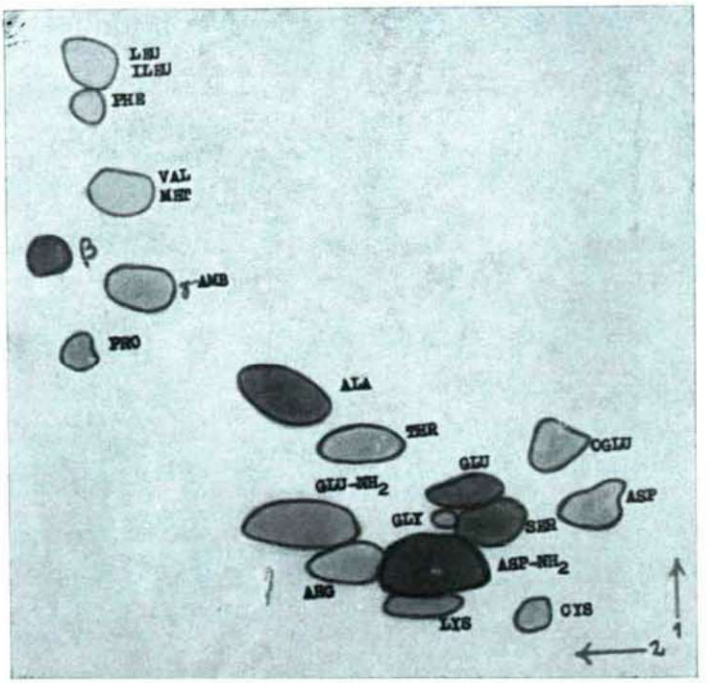

Fig. 4. Chromatogram of the cluster extract of rice with abundant N-supply. Dunghan shali, blooming. „ß": blue spot.

Two-dimensional chromatograms were also prepared from the different parts of rice and of these we show in Figs. 3 and 4 the chromatograms of the leaf and cluster extracts. It is true, that from two-dimensional chromatograms the quantitative deductions are less valid, however, it may be considered that there was a much smaller blue spot in case of the cluster extracts than in that of leaf-extracts. This fact seems to be supported by one-dimensional determinations of the eluted spots, too. Asparagine yields a strikingly large spot in the two-dimensional chromatogram of the cluster, and also the glutamine spot is considerable. In the chromatograms of the cluster there appeared, above aspartice acid, oxyglutamic acid, as well, the demonstration of which is rather difficult.

The one-dimensional chromatogram in Fig. 5 was made with hydrolysed samples. In the free amino acid extracts of rice supplied with large amount of N before sowing there is a large blue spot. Leaves purified from the free amino acids do not containt the blue substance in their hydrolysate. In the chromatogram of hydrolysed free amino acid extracts the blue spot appeared unchanged.

Fig. 6 shows that the blue spot refers to disturbances not only in the case of the Hungarian-bred Dunghan shali but with the Soviet sorts, as well. Provoked by heavy N-supply, the blue substance appeared in each of the Soviet sorts studied by us, however, in an amount smaller than in the Hungarian sorts.

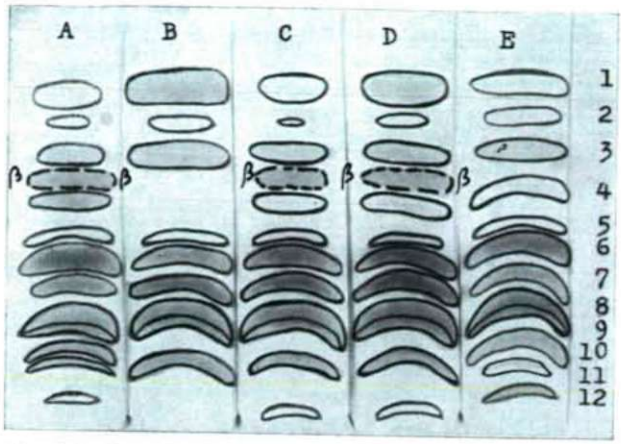

Fig. 5. Amino acids of leaf extracts and hydrolysed leaf proteins of rice (Dunghan shali) in blooming. Supplied with large amount of N. „A": free amino acids. „B" hydrolyzed leaf protein purified from free amino acids. „C" and „D": hydrolyzed free amino acid extract, simple and double amount. „E": universal standard with 50 μg total amino acid content. Number as in Fig. 1.

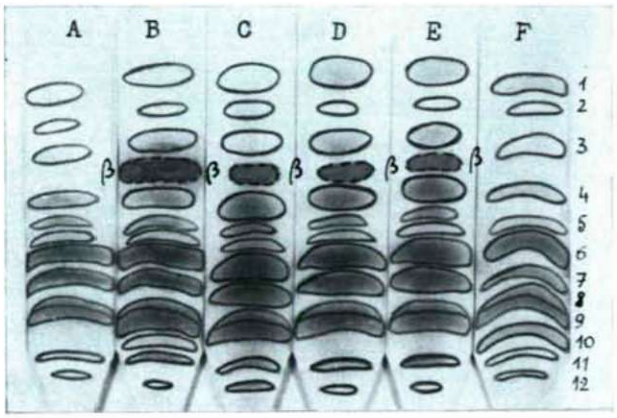

Fig. 6. Chromatograms of rice leaves, Hungarian and Soviet sorts. Blooming. „A" : Dunghan shali without fertilizer. „B"; Dunghan shali with abundant N-supply. „C": Dubovskiy 129 with large amount of N. „D": Krasnodarskiy 424 with large amount of N. „E": Krasnoarmenskiy 313 with large amount of N. „F": universal standard with 50 μg total amino acid. „ß": blue spot. Numbers as in Fig. 1.

At the same time no blue spot was demonstrated in the plants without a fertilizer.

In Fig. 7 the leaf-extracts of three Hungarian-bred and three Soviet-bred rices are compared. It can be seen that in the one-dimensional chromatogram developed in phenol-water solvent the blue spot framed with broken line showed the highest Rf value. From the chromatogram it appears that in the Hungarian

sorts supplied with great amount of N the blue substance is more abundant than in the Soviet-bred rice supplied with a similar fertilizer. Several hundred chromatograms were prepared and the result was always the same. In the control of the Hungarian Dunghan shali there was no blue substance or it appeared only in traces. Diseased samples, however, occurred in the control, as well, probably due to the unfavourable weather conditions.

Fig 7 shows that the blue spot appears starting both from the fresh substance and from the dry one. Thus the blue substance is not a decomposition product formed during drying.

In each of the three years of investigation the blue substance could be demonstrated in the lower leaf storeys of rice plants, but in an amount less than in the upper storeys.

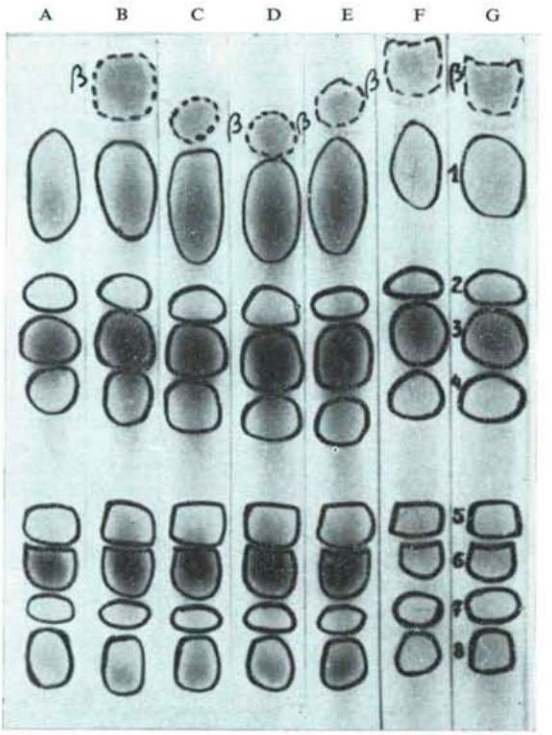

Fig. 7. Chromatograms of leaf extracts of Hungarian and Soviet rice sorts developed with phenol-water solvent. First strip on the left is the extract of unfertilized control, others of that supplied with large amounts of N. The left five strips are made from fresh substance, the two ones on the right from dry substance. „A": and „B": Dunghan shali. „C": Krasnoarmenskij 313. „D": Dubovskij 129. „E": Krasnodarskij 424. „F": Szarvasi 63. „G": Kákai 203.

1. Phe, Leu, Val
2. „-Amb.
3. Glu-NH_2, Ala, Arg
4. Thr, Lys
5. Asp-NH_2
6. Ser, Glu
7. Cys
8. Asp

„ß"-blue spot

In Fig. 8 one can see the clusters with barren flowers of rice attacked by brusone, partly or wholly whitened. The leaves of these plants are green but with brownish-grey spots. As to yield, we must note that in case of Dunghan shali, with abundant fertilizer before sowing, the decrease of yield ihad been of about 80 p. c. with other Hungarian sorts 30—70 p. c. and there was no decrease in the yield of the Soviet-bred rices.

The rice plants of the State Plantation, Pale were not unilaterally supplied with N before sowing, yet, several diseased spots appeared the previous year. The plants of the diseased spots contained a considerable amount of blue substance, but the healthy ones did not. Thus only plants changed by the unilateral N-supply give the blue spot but so does rice diseased in other ways, too.

Our experiments showed that, the unknown substances does not give the alloxane and foline reactions of the amino acids, it is not decomposed by L-amino acid oxydaze (information from G. FARKAS), therefore its amino acid character is doubtful.

Fig. 8. Barren clusters of diseased Dunghan shali supplied with a great amount of N. They are partly or wholly whitened.

Discussion

Dunghan shali with abundant supply of N before sowing has very loose tissues and is susceptible to diseases. Such an abnormal physiological state can equally be caused by prolonged cool weather, lack of sunshine, cold soil or flooding water or by lack of phosphorus. The blue spot, first demonstrated by us (PÁLFI 1964a) refers to such a weakened state and probably disturbances in the protein metabolism. The blue substance can be demonstrated in any part of such a rice plant, although in different amounts — even from apparently healthy plants. The upper leaves of rice, that are still green at the complete ripening, contain the blue substance only in traces, and in such leaves the protein synthesis is already rather weakened.

During the first two years only Dunghan shali was investigated. In the third year beside it six Hungarian and five Soviet rice sorts were included.

The results show that in the leaf extracts of plants abundantly supplied with N or of plants diseased in brusone the blue spot could be demonstrated in all of the twelve sorts. On the contrary, it cannot be demonstrated in the case of the healthy controls. The „blue substance" content of the Soviet sorts is much less than that of the Hungarian ones. The brusone susceptibility of the Soviet sorts is much less, as well. This is supported by last year's results, since being provoked, all of the Hungarian sorts became more or less contaminated by *Piricularia* in contrast to the Soviet sorts. It must be mentioned, however, that Dunghan shali, having nice long grains, is far the best of all the studied sorts as regards to nutritional value, physical and cuisine properties, as well.

The blue substance had been demonstrated from each leaf storey of the plants but to a less extent from the lower leaves. This is in agreement with our former statement (PÁLFI 1963, 1964 b, 1965 a) that free amino acids, as well, can be found in different concentrations in the different leaf storeys and that the free amino acid content of the leaves is the less in the lowest storey. The same had been established by FRENYÓ (1961) in respect to inorganic nutrients of rice. In the leaves of the diseased rice not only the blue substance appears in large amounts but free amino acids and amides, too.

In the experiments with wheat diseased with mycosis FARKAS (1963) and others with different plants (FELDERMANN and HANKS 1964, GRINEVA 1964, SEHGAL and BOONE 1964) obtained similar results.

Summary

1. During paper chromatographic analysis of amino acids of rice leaves growing in the field an unknown substance had been demonstrated. This substance („blue substance") after being treated with ninhydrine and a following copper-salt fixation gives a blue spot instead of a red one, characteristic of the amino acids.

2. Investigations of samples collected in three years showed that the appearence of the blue substance indicates an abnormal physiological state, probably a disturbance in the amino acid and protein metabolism.

3. The blue substance occurs not only in the leaves but in the root, stem and cluster of rice, as well. It was found in the greatest amount in the leaves and the stem, less in the roots and the least in the cluster.

4. The blue substance content of the single leaf-storeys of rice plants is different: there is less of it in the lower leaves than in the upper, younger ones. Thus the blue substance appears in a larger quantity where the protein metabolism is the most intensive. At total ripening of the grains, when the protein metabolism decreases even in the upper green leaves still alive, no blue substance could be demonstrated there.

5. The demonstration of the blue substance had been performed on plants grown under varied conditions: in cool weather, artificially performed lack of sunshine, *Piricularia* infection and addition of abundant N-fertilizers. Results obtained in case of six Hungarian and five Soviet sorts prove that the blue-test can be applied for the demonstration metabolic disturbances (probably disturbances of the protein metabolism) of rice.

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LIGHT-INDUCED CHANGES OF VACUOLAR CONTRACTION OF SENSITIZED CILIATA

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It is known that some light absorbing materials, dyes photosensitize the various organism, or rather their protoplasm (Blum, 1941). Many enzymes, plasma proteins of the sensitized organisms may become denaturalized, and what is especially frequent with viruses, bacteria and protozoa, they may quickly die. The phenomenon called photodynam (Tappeiner and Jodlbauer, 1904) reveals important plasmic changes and is therefore a useful means in getting acquainted with the biology, the physicalchemical behaviour of the plasma. This is why it semed important to examine the photodynamic influence of known dyes on the frequency of contractile vacuoles in the *Platyophrya lata*.

Material and method

The ciliate *Platyophrya lata* Kahl breeding in root extract were made to fast after centrifugation in a solution of a determined ion composition (Biczók, 1961), then they were dyed with a partly purified solution of 1:100 000 dilution of fluorescein, eosin, rhodamine B, rose bengale, methylene blue, toluidine blue, trypaflavin, neutral red, Janus green B and auramin O controlled with measurements of absorption-spectrum. The isolated specimen was lit with the 500 or 25 000 lux strong light of a 15 W and 6 V lamp. We tried to eliminate the heat effect of the light by placing a thin $CuSO_4$ solution between the light source and the hanging drop. On the average we examined 25 specimens each time. After reading off 10 frequencies in each case we inserted a pausa of 5 minutes.

Result, discussion

We expected a major effect from the xanthene dyes among the sensitizing agents because they are well inducible, their photodynamic effect, their photooxidation are known to be great (Hertel, 1906; Jodlbauer and Haffner, 1921; Gilbert, 1942; Lábos, 1966a, b). Besides this Hyman and Howland (1940) have demonstrated of two members of these dyes, that injected into an *Amoeba*, they caused over-activity of the vacuoles. Two dyes, eosin and rose bengale have since then been in the highlight of interest.

Our results are shown in the table. The measured results, the averages refer to the function of vacuoles contracting in intervals shorter than 20 sec. We have

expressed the vacuolar frequency increase and decrease not only in % but also by the quotient of the average sums of the vacuolar frequency of the animals before and after dying. Evaluation was difficult because part of the sensitized animals encysted (Table). Encystment changes the normal rhytm of vacuolar pulsation before the rotating movement accompanying encystment. When the *Platyophrya* died before rotation we could not explain the fluctuation in the contraction of the vacuole. Furthermore certain dyes make their toxic influence felt even in a 1:100 000 dilution which is not identical with the damaging effect resulting from photooxidation. We think here of methylene blue, toluidin blue and Janus green B.

Taking all this into consideration the following may be stated:

1. In the majority of cases vacuolar pulsation becomes accelerated in the photosensitized animals as compared to the undyed animals. In respect of this effect the following order may be established in the xanthene dyes:

eosin > rose bengale > rhodamine B > fluorescein.

The Janus green B belonging to the mono-azo group has proved to be a very effective sensitizer; it increases vacuolar frequency by a large percentage and reacts most intensively to a stronger light. The difference between the vacuolar frequency of colourless and dyed animals functioning with decreasing tendency under the influence of light.

The photodynamic effect of neutral red an auramin O is moderate, although at 25 000 lux the quickening of the vacuolar frequency is considerable with these too.

2. The increase of light intensity caused in its total average an increase in the number of contractions with all sensitizers. As it was to be expected, the reaction of the xanthene dyes is most conspicuous in this respect also. Photoreaction at 25 000 lux took place in almost the same order of the dyes as at 500 lux.

3. The smaller part of the sensitized *Platyophryae* (also the undyed ones) responded to light with the slowing down of the vacuolar contractions. Differently from the other indexes this opposed tendency is expressed by values under 1. In my opinion these indexes have rather a precarious value in appreciating the phenomenon of photodynam since they express the frequency of decreasingly, weakly functioning vacuoles. In spite of this fact it is conspicuous, that the indexes referring to xanthene dyes do not exceed an average of 0,8, while the thyaziner are all below their value, i. e. the intervals of contraction frequency in the undyed state and those occuring at 500 lux after dying are considerably greater here.

4. Some degree of regularity may be detected the frequencies of the unchanged, quickening and slowing contractions shown in brackets. In eahc series of experiments the number of vacuolar frequency of undyed, then sensitized specimens under the influence of ligth was as follows:

unchanged < quickening > slowing,

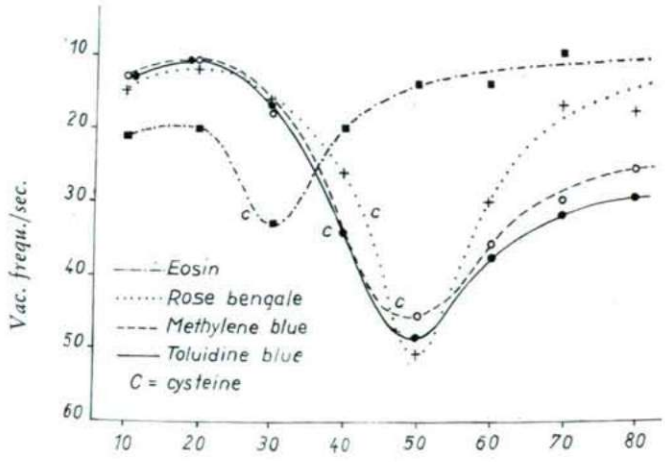
i. e. under the influence of light the vacuoles contracting at slower frequencies become quicker, those functioning quickly do not change or rather slow down.

5. Under the influence of light the vacuolar rfequency of the photosensitized animals changed, usually becoming quicker. Depending on the quality of the

Table

Dyes	Frequency of vacuole contractions						Encystment during examination
	after dying at 500 lux (Average frequencies in brackets)			index showing ratio of original state and change after dying		index showing ratio of change after dying at 500 and 25 000 lux respectively	
	unchanged	faster	slower	increase of frequency	decrease of frequency		
	%	%	%				
Eosin	5 (11)	75 (14,6 / 10,8)	20 (10,3 / 12,3)	1,352	0,837	1,408	8
Rose bengale	15,4 (12,5)	69,2 (14,4 / 11,2)	15,4 (13 / 16)	1,286	0,813	—	3
Rhodamine B	5 (15)	60 (15,5 / 12,8)	35 (11,4 / 13,6)	1,211	0,838	1,500	2
Fluorescein	21,1 (13,5)	52,6 (14,2 / 12,6)	26,3 (15,4 / 19,2)	1,119	0,802	1,416	6
Methylene blue	9,5 (8,5)	76,2 (15,5 / 11,6)	14,3 (19,3 / 32,3)	1,336	0,597	1,408	—
Toluidine blue	14,8 (11)	51,9 (13,1 / 10,4)	33,3 (13,7 / 24,3)	1,250	0,564	1,323	—
Trypaflavin	19,1 (10)	47,6 (14 / 11,6)	33,3 (11,3 / 15,9)	1,207	0,711	1,296	9
Janus green B	9,5 (12)	71,4 (15,6 / 11,5)	19,1 (14 / 25,8)	1,357	0,543	1,525	—
Neutral red	26,1 (10)	30,4 (16,9 / 13,9)	43,5 (12,1 / 16)	1,216	0,756	1,385	1
Auramin O	5,3 (10)	47,35 (15,4 / 12,6)	47,35 (12,3 / 13,9)	1,222	0,885	1,376	4
Colourless (at 500 and 25,000 lux)	29,9 (12,6)	39,3 (14,1 / 12,9)	30,8 (13,1 / 14,1)	1,093	0,929	—	3

dye and the intensity of the light the acceleration decreased after 10—15 minutes, sometimes after a longer period and ceased almost simultaneously with the loss of the ability of locomotion. With some dyes this condition occured after long hours (e. g. with fluorescein, trypaflavin, auramin O). We may have here a case of sensitized photooxidation which may inactivize DNA (SIMON and HELEN VAN VUNAKIS, 1946; SUSSENBACH and BERENDS, 1964), important enzymes (TAPPEINER and JODLBAUER, 1904; RAPOPORT and co-workers, 1965), important structure-forming, SH-groups containing compounds (WITTNER, 1957; WACKER and co-workers, 1963; RENSBURG and co-workers, 1965). Many antioxidants, cystein among them, give effective protection against this damaging influence. Therefore we made an attempt to reactivate animals sensitized with the enumerated dyes and not yet noticeably damaged by light. We gave cystein (15—25 mM) to the weakly moving specimen with slow vacuolar pulsation. Those *Platyophryae* in the protoplasm of which there were already vesicles distinguishable, colescent vacuoles, they died quickly. Otherwise ciliar motion and vacuolar activity became quicker (Graph). The reaction shows, that the SH-groups which are oxidated during the effect of photodynamism play an important part in the function of the *cilia* contractile vacuoles.



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COMPARATIVE CYTOLOGICAL EXAMINATIONS OF THE PARAVERTEBRAL GANGLIA OF TADPOLES AND FULL-GROWN FROGS

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The light- and electronmicroscopic examinations have resulted in a more perfect cognition of the structure of nerve cells. Already in the early years of the century, the research workers observed granules of different size in the neurons, even without staining. These granules were classified into two groups: the fine granuler lipofuscin pigment stained yellowish-brown was classed into one of the groups, and the rough granular melanin pigment of brownish-black colour into the other one.

The opinion of scientists concerning the occurrence of pigments is not uniform. According to some of them, melanin is restricted but to certain cell groups of the central nervous system *(Nucleus niger, Locus coeruleus, nucleus of the dorsalis vagus);* in other *opinions* (KUNTZ, 1934; BARGMANN, 1948; STRONG and ELWYN, 1948, and LARSELL, 1951), it occurs in men also in the spinal and sympathetic *ganglia.* The latter opinion is not confirmed by the recent examinations of SULKIN (1953). According to him, the dark staining in the sympathetic cells is caused by lipofuscin present there, the chemical nature of which differs strongly from that of melanin.

About the chemistry of pigments we know but little. BIELSCHOWSKY (1928) and LEHNARTZ (1942) supposed that in the nerve cells melanin developes through the oxidation of dihydroxiphenilalanin (DOPA) and o-quinon differring, anyhow, substantially from the melanin of skin (LE GROS CLARK, 1945).

The yellowish-brown lipofuscin granules arise, however, in the opinion of LILLIE (1948) and PEARSE (1953), in the way of a progressive oxidation from a precursor of lipid content. Pigment substances, thus also the *„ceroid"* pigment in liver, are considered by PEARSE as middle products of oxidation processes. And SPIEGEL and ADOLF (1822) try to derive the yellow pigment from the black one.

Completing the light microscopic pigment examinations with electronmicroscopic ones, we may conclude that the imbedded substances of the vegetative nerve cells, or at least a part of them, can be classed into the group of lysosomas showing an acid phosphatase activity, changing, growing, and achieving possibly the size of more microns (TAXI, 1965). The identity of the genesis of lipofuscin and lysosoma is proved by a series of experiments verifying that the lysosoma pigment transforms, under the influence of a radioactive irradiation, into lipofuscin turning, however, back into lysosoma after the irradiation that stopped, again showing an acid phosphatase activity.

Material and method

I have examined the species *Pelobates fuscus fuscus, Rana ridibunda,* and *Bufo viridis* in different states of their development. The *ganglia* of the limiting fascicle of one side, dissected from the animals, have been fixed in a pH 7,4 osmium solution pufferred according to MILLONIG; and the *ganglia* of the limiting fas-

cicle of the other side in formalin. The *ganglia* fixed for the electronmicroscopic examinations were embedded into Araldit and the slides examined by an electronmicroscope TESLA 242 D. The histochemical demonstration of pigments took place with SCHMORL's method. In addition, also stainings with Sudan black, alkali tetrasolium, Toluidin-blue, Gallo-cyanin, Acridin-orange, Janus-green, and neutralred were carried out for demonstrating collectively the other *organella* of cell, as well.

Examination of the ganglia of the sympathetic limiting fascicle

I have examined the ontogenesis of the vegetative nervous system of tadpoles for several years. At the tadpoles of different development I have followed with attention the formation of *ganglia* of the *truncus sympathicus* (HORVÁTH, 1965): from the sympathicoblast through the sympathicocyte till the fully developed *ganglion*. The endoamitotical division of the sympathicoblast cells can well be observed by a light microscope. The 4—7 μ thick slides of the vegetative *ganglia*, prepared in series, were treated parallel with reagents and stains of different pH. From the methods used by me for demonstrating lipofuscin, although not specific ones, compared with literary data known by me, the conclusion can be drawn that the pigments observed belong to the group of lipofuscin. That result is confirmed also by the electronmicroscopic examinations where I could observe in some of them, apart from the polynucleosity of the sympathicoblast cells, some pigments of different size, stained well by osmium. Considering that the research objects were tadpoles in different states of their development, these results may supposedly serve as a basis for the genesis of lipofuscins, *resp.* lysosomalike cell components unknown, as yet. In the course of my further examinations, I should like to clear up approximatively the G. E. R. L. functional system set up by NOVIKOFF in 1964, trying to discover a connection between GOLGI's apparatus, the endoplasmatic *reticulum*, and lysosoma. In my present paper I want to get some conclusions concerning the time of appearance of the pigment and its role inside the cells.

The sympathicoblasts deriving from the neural crest form smaller or bigger groups in the line of the limiting fascicle, suitable for an endomitotic division. The sympathicocytes produced in the course of division contain, in contradiction to the giant sympathicoblasts, not more than two *nuclei* and but a single appendix. In this way, the sympathicocytes can be divided once more. The lipofuscin pigments can be observed in a great quantity first of all in the sympathicoblasts of 24—32 μ capable of being divided endoamitotically more times (Fig. 1). And also the fusion of these granules of 0,01—0,5 μ can be observed. The granules can supposedly fuse and, surrounded by a simple membrane, turn into lysosomas (Fig. 2). In the other ganglia of the more developed tadpoles the amount of lipofuscin pigments falls to a minimal one and, in the same time, in a bigger amount, also the storage of *mitochondria* may be observed, apart from the tigroid granules. This picture can be seen in the *ganglia* of the limiting fascicles of one year old frogs (Figs. 3—4). In the *ganglia* of older animals, at

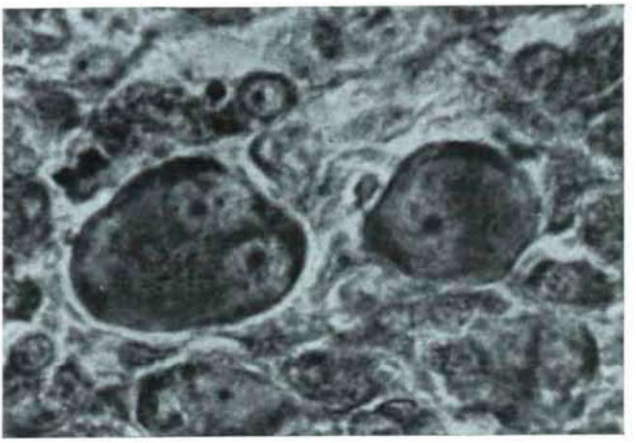

Fig. 1. *Pelobates fuscus* (tadpole): Large amount of lipofuscin granules in the cytoplasm of the sympathicoblast dividing endoamitotically. SCHMORL's method. Microphotograph, x 675.

an advanced degerenation, the cells perish after a major increase of the tigroid granules (Fig. 5). In these cases the *nucleus* of excentric site shrinks considerably, and a number of lipofuscin granules take place, in a group, at the pole facing the *nucleus*.

From the above-mentioned accumulation of a major amount of pigments in the giant sympathicoblast cells of tadpoles, and from the occurrence of a comparatively smaller number of pigments in the *ganglia* of the fully developed frogs the conclusion may be drawn that the biochemical processes in cells must have been carried out, even if in a lower degree, supposedly in the presence of other *organella* producing and storing energy. The proportion of the comparative amounts of tigroid granules and *mitochondria* is worth mentioning, as well. In the degenerating sympathicoblast cells, even in spite of the high degree of protein synthesis, the number of *mitochondria* is very low compared with that of the fully developed *ganglia*. The quantitative difference of tigroid granules may change in a high degree depending upon the functional state, therefore, I don't want to make any comparison in this field.

After the conclusions made known above I have to mention some literary data, as well, concerning the intracellular role of pigments. It is generally accepted that the amount of yellow pigments increases with the age, at a lasting output, and in the case of some diseases. Yet we can find several ideas opposite to that statement. According to KUNTZ (1932) the pigmentation of human vegetative nerve cells may be observed in the middle period of his life without influencing by it the cell productivity. WHITE (1887, 1889) asserts, alluding to animals on a lower degree of phylogenesis, that the vegetative cells are not pigmented, as yet. The pigments appearing later result in a sure decrease of functioning. SCHAEFER (1893) considers the pigmentation not as a degenerative phenomenon but as a function-increasing one. According to HYDÉN (1950),

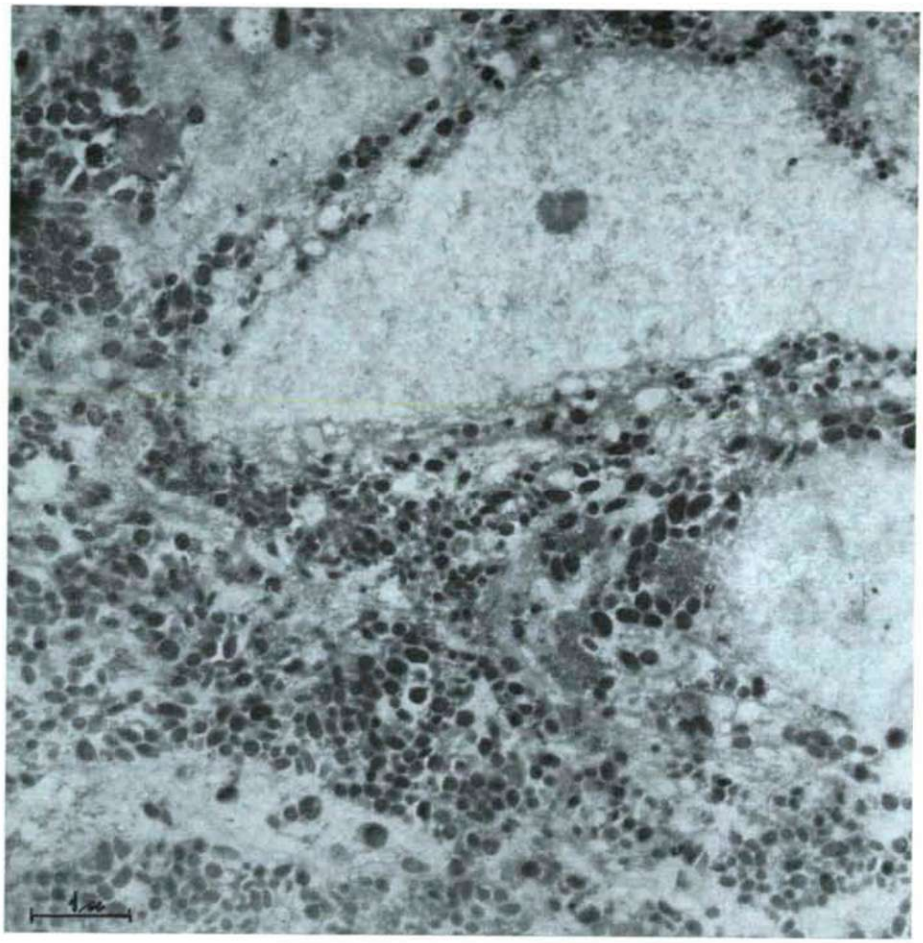

Fig. 2. *Pelobates fuscus* (tadpole): An electronmicroscopic photograph about a binuclear cell of a sympathicoblast in which scattered lipofuscin granules are taking place. Araldit embedding. Magnified x 15 050.

after the yellow pigments had appeared, the cells become differentiated, as proved also by the presence of the cytoplasma nucleotide demonstrated beside the pigments. HYDÉN and LINDSTRÖM (1950) assert also on the basis of their examinations concerning the mass determination by roentgendiffraction that the pigment accumulation at the advancing age results in a further chemical organization of neurons. It may be supposed on the basis of the histochemical examinations of GEDIGK and BRONTKE (1956), as well, that the lipopigment plays a peculiar role in cell metabolism. We must doubtless attribute a considerable role to the pigments in the synthesis of the high degree metabolism taking place in the sympathicoblasts of tadpoles.

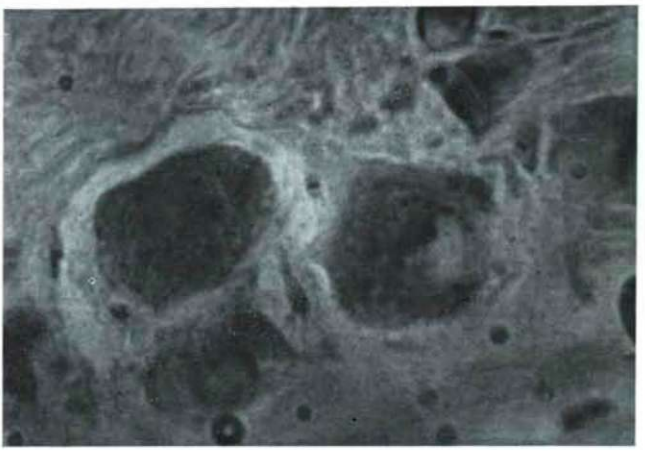

Fig. 3. *Bufo viridis:* Tigroid granules in the nerve cells of *Ggl. sympathicum* VI. Toluidin-blue staining. Microphotograph, x 675.

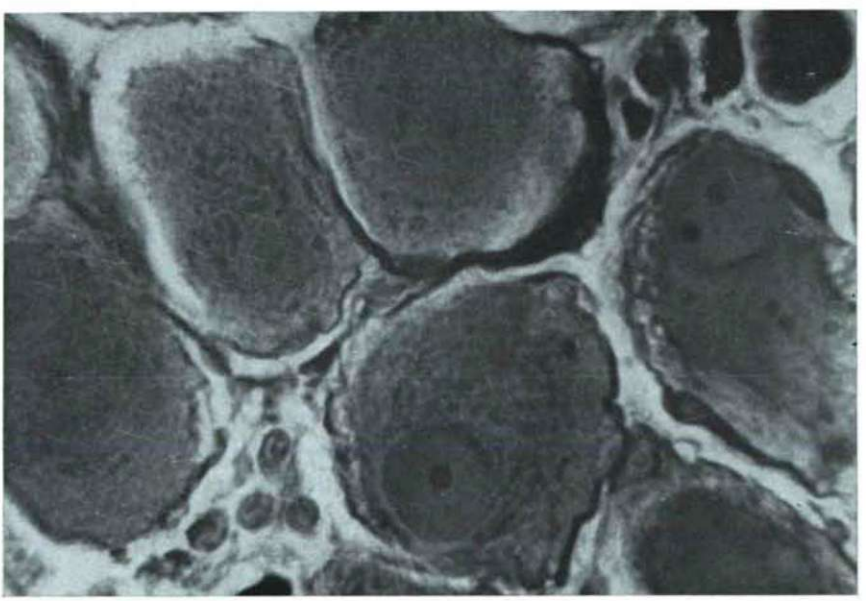

Fig. 4. *Rana ridibunda:* Demonstration of *mitochondrium* in the *ganglia* of *Ggl. sympathicum* VI. Stained by Janus-green B. Microphotograph, x 1054.

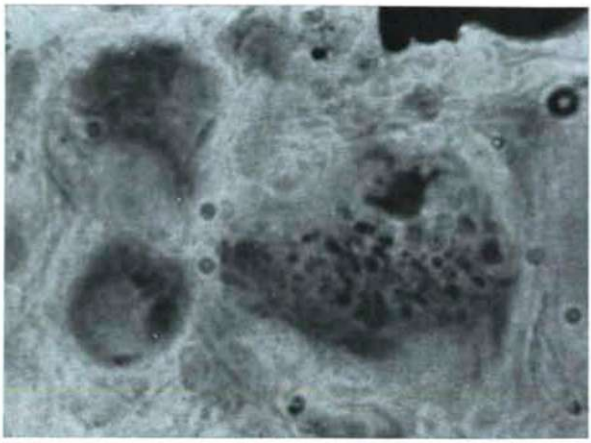

Fig. 5. *Rana ridibunda:* Nissl substance accumulated in the degenerating nerve cells of an old frog. (From *Ggl. sympath.* VI). Gallocyanin-chrome-aluminous method. Microphotograph, x 675.

There are differing opinions concerning the genesis of the pigments of nerve cells, as well. DOLLEY (1917) derives the pigment from the nucleic subtance. According to MONROY (1934/35), however, the granules of pigment develop from the *mitochondria.* Therefore, several researchers are occupied by clearing the raised problems. Partly I should transgress the framework of this paper, partly also I should be constrained to build upon mere suppositions if I tried to answer these important questions.

Summary

I can summarize the results of my comparative cytological examinations carried out on tadpoles of different development *(Rana ridibunda, Pelobates fuscus fuscus, Bufo viridis)* and on fully developed frogs, as follows.

1. I could demonstrate a large amount of lipopigments taking sporadically place in the giant sympathicoblast cells of tadpoles in development.

2. The amount of lipopigments decreases gradually from the young neurocytes till the full-grown nerve cells, instead of them an increase of the number of the tigroid granules and that of *mitochondria* my be observed.

3. In the vegetative *ganglia* of older animals the lipofuscin granules occur forming lesser groups in the cytoplasm opposed to the *nucleus.*

4. In my opinion, the large amount of the pigment granules of the sympathicoblast cells may have an absolutely important role in cell metabolism.

5. We may conclude the degeneration of nerve cells not so much from the pigment increase but more from the inrease of the number of tigroid granules and from the decrease of that of *mitochondria.*

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THE MICROSCOPIC INNERVATION OF THE GILL-APPARATE IN SCORPAENA PORCUS

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The anatomical and structural difference of the gill-apparate in fishes was examined by several authors as it is proved by RAUTHER's monographical work (1937). Especially many data were given by BEVELANDER (1935) about the structure and function of their epithelial cells. Well-known examinations threw light upon the physiology of the gills (LUTZ, WYMAN 1932, INVING, SOLANDT 1935, COPELAND 1948) However, the problem of gill-innervation, mainly in microscopical respect, except DE BOYD's work (1936) may be considered uncleared. Concerning the innervation of the gills was determined in general that the first branchial arches are supplied with glossopharyngeal nerves and from 2—4 with vagal ones (BÜTSCHLI, ROMER, WIEDERSHEIM). There are only suppositions about the participation of the trigeminal, facial nerves and of the sympathetic system in the innervation of the gill-apparate.

Material and method

We studied the innervation of the gills in the *Scorpaena porcus*. This animal was preferred because we could get it in great number due to the kind help of the scientifical workers in the Research Institute of Split. On the other hand the function of their gills is usual and very surprising. They swim quickly in the bottom of the aquarium and stop many times, open the opercular apparate and move the gills. The moving became stronger lifting the animals. Under dry circumstances the skin of the head and the palatinal membrane became pinky, likely owing to the change of the blood circulation and breathing, and about 5 minutes the gills move regularly.

The gill apparate were fixed in 10 p. c. formaldehyde. The anatomical section was prepared with the *truncus arteriosus*. The microscopical slides were made with frozen microtome separating first the gill-lamellae, and the impregnation with BIELSCHOWSKY—ÁBRAHÁM's method. The thickness of the slides was 20—25 μ.

Different parts in the gill apparate and their innervation

The gill apparate has 4 well developed double branchial arches with two lamellae in *Scorpaena porcus*. The anterior ends of the arches came together in the *entoglossum* whereon the lingual part is formed. The posterior ends are connected with the *pharynx*. The whole apparate forms a unity with the hyoi-

dean hemibranch, with the mucous membrane of the oral cave and with the musculature belonging to the branchial, hyoideal and pharyngeal region.

The two lamellae are placed to outward side of the osseous branchial arch. They are situated on the concave pharyngeal margin of the branchial arch merged into one another at the basis. Opposite the gill-lamellae at the inward side, the branchial arch is fringed with bony processes, so-called gill-rakers. They are covered with the mucous membrane. This is continuation of that in the oral cave and spreads to the branchial arch too, forming the connection between the gill-lamellae and gill-rakers as well as the loose connective tissue around the branchial arch.

So, the above mentioned parts including their special and rich blood circulation and musculature, show difference of each other, not only structurally but also in respect of innervation.

Gill – lamellae

The structure of the gill-lamellae as common in the osseous fishes, consists of filaments of different size. They are longest at the middle and they gradually decrease towards the ends. In the central part of the filaments, in the loose connective tissue cartilagineous trabecle may be seen in right angles to the osseous branchial arch. Here are the arteries and veins, originating from the branchial vessels, running parallel to the supporting cartilage of filaments. Sometimes at the basis of filaments striated muscle fibres appear too. The entire surface of each of the filaments is covered with respiratory epithelial cells. Many delicate indentments to fine little filaments grow their respiratoric surface.

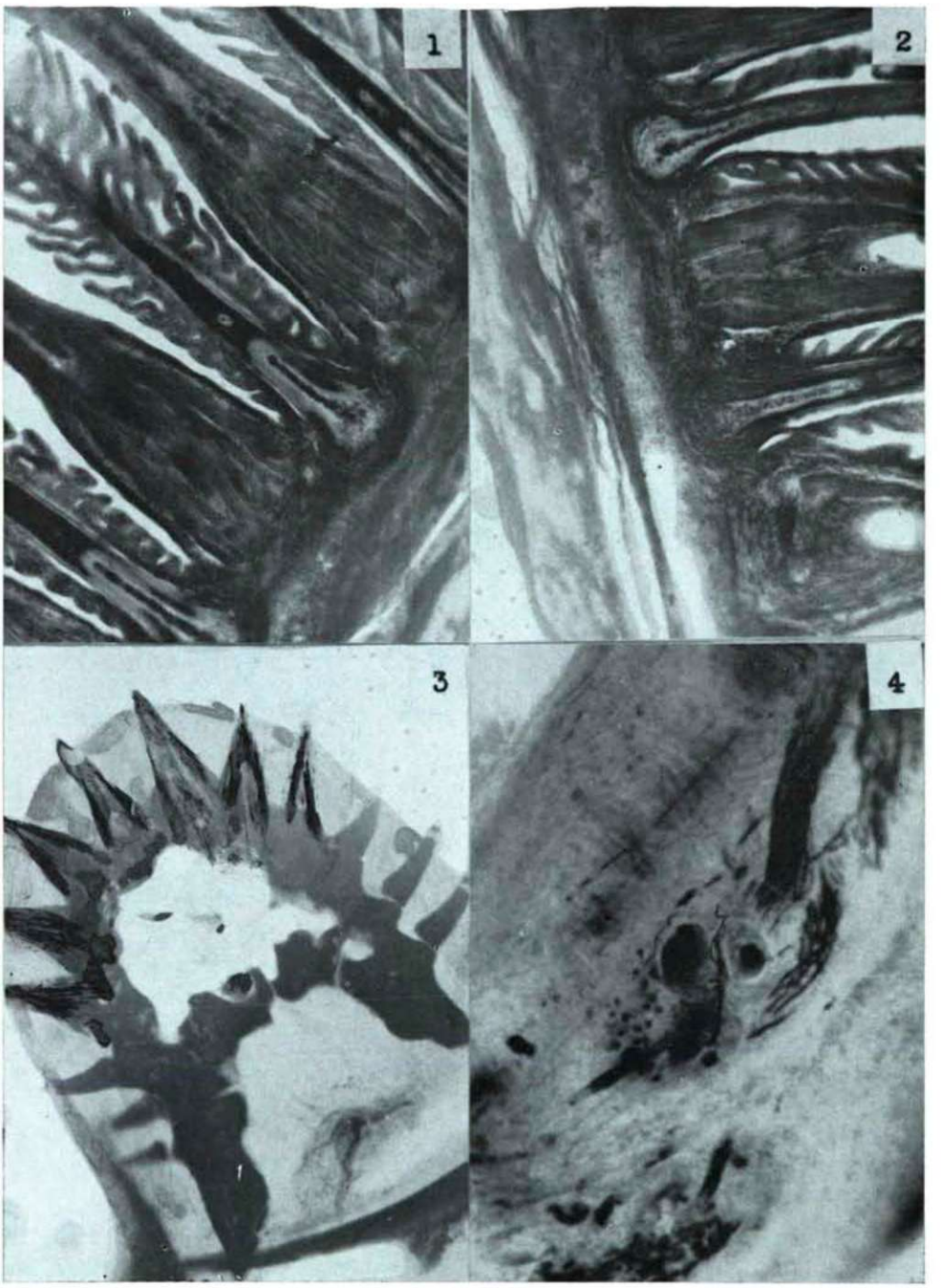
The gill-filaments of *Scorpaena porcus* have always broad base and show two types. One type of the filaments possesses supporting cartilage the wide diameter of their base originates from the large devided bulb. The other type shows surprising many striated muscle fibres, very large blood vessels and abundant connective tissue at the basal part. Their decrease caused the thinning of the filaments close to the top. The two kinds of filaments is regularly alternating (Table I. Fig. 1). At the endings of the lamellae the filaments show an absolute lack or very small cartilage and in most of the cases the muscle fibres do not occur either. These pictures are in full agreement with the description of the gill filaments of fresh water fishes by other authors (KRAUSE 1921, RAUTHER 1925, SCHÖTTLE 1910).

The nerves of the filaments reach the base in one trunk. They come from the connective tissue around the osseous arch. Here the main trunk is found in a close connection with the branchial artery and vein (Table I. Fig. 2). The nerves have many thick fibres forming coils in their way and quite thin fibres running

Table I.

Fig. 1. Alternating structure of gill filaments.
Fig. 2. Blood vessels and nerve trunks connections to the filaments.
Fig. 3. The thorns of a gill-tooth.
Fig. 4. Nerve cells in the arterial *plexus*.

TABLE I



always straight. Both, however more frequently the thicker ones have varicosities. The lateral branch of this trunk, the so-called filamentary nerve, runs obliquely to the filaments and forms a very rich *plexus* in the basal part.

Alternative change can be seen not only in the structure of filaments but also in the innervation. The supporting filaments are always poor of nerves but the others are so richly supplied that is a rarity (Fig. 1). Three kinds of arrange-

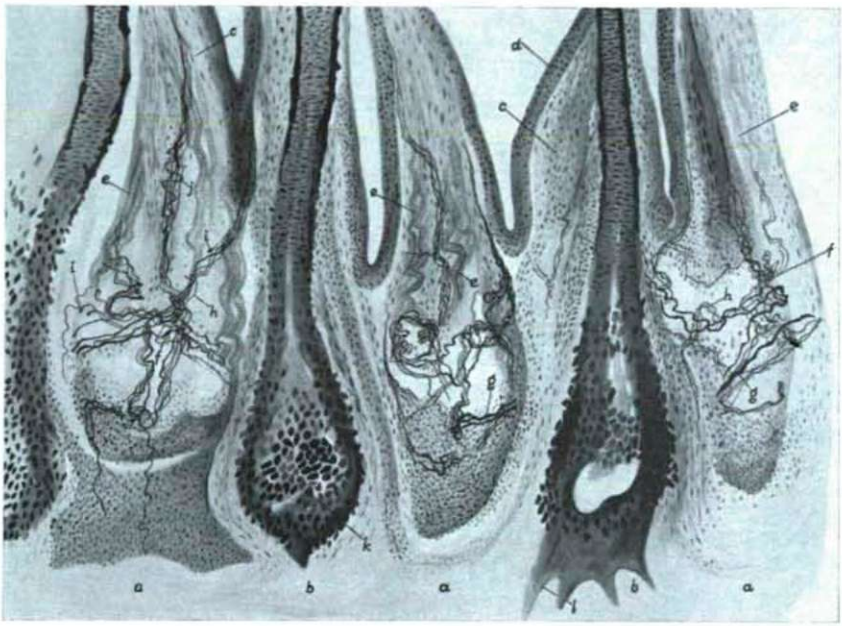


Fig. 1. *Scorpaena porcus:* Alternating structure and innervation in the basal parts of the gill filaments in the middle of branchial lamellae. a — bases of the moving and vascularised filaments, b — bases of supporting filaments, c— connective tissue, d — respiratory epithel, e — striated muscle fibre, f — ring *plexus,* g — thick nerve fibre, h — thin nerve fibre, i — nerve ending, j — spiral fibre, k — cartilage, l — thorns of the gill raker. BIELSCHOWSKY—ÁBRAHÁM's method. Magn. 120x.

ments could be separated in the dense *plexus* at the base of the filament. One is quite superficial and shows ring form. A part of the fibres runs to the striated muscle fibres and an other one to the filamentary artery. We suppose most of the thick fibres are connected with the musculature. The filamentary muscle fibres get very rich nerve supply. Their special innervation will be described in the chapter of „The muscles of the gill apparate".

Some of the fibres originating from the ring *plexus* run to the filamentary arteries and form very rich and dense plexuses in the arterial wall together with the fibres coming here in the *adventitia* of the vessels and in all probability belong

to the cranial sympathetic system. Sometimes little black end-spots or rings could be seen at the endings of the nerve fibres, especially in the *media* and larger end-plates with neurofibrillar structure in the *adventitia*. The former must be the endings of the *effector* fibres and the latter of the receptors.

May be that some of the free end-fibres of the plexuses run quite close to the epithelial cells accumulated between the two filaments but intimate connection was never visible. Some of the fibres of the plexuses run upwards along the filamentary arteries. Many spiral fibres appeared among them (Fig. 1, j). However, we would emphasise that the top fibres originating from the plexuses reach only the middle of the filaments. No fibres could be found in the upper part of the filaments where the respiratory epithelial cells and the capillary system were dominant. So the function of the respiratory cells is similar to that in the lungs of higher vertebrates free of close nerve connection.

Gill – rakers

The gill-rakers are seemingly very simple and insignificant formation of the gill apparate but according to their structure and innervation they must play an important role in the life-function, especially in the feeding of the *Scorpaena porcus*. Taking into consideration their structure they consist of separated hillocks at the opposite ends of the supporting gill-filaments. Structurally they are similar to the teeth of the higher vertebrates and probably have the function like that of the teeth, as the real maxillar and mandibular teeth are good enough to seize the food but not so strong to chew it. So it is right to call them gill-teeth. The main substance of the gill-teeth is compact bone forming 8—16 thorns towards the oral cave and *septa* of different sized to the *pulpa* cave. Outside they are covered with the mucous membrane but the ends of the thorns run through most of the cases the membrane (Table I. Fig. 3).

Inside of the gill teeth the connective tissue full of blood vessels and nerve fibres is found. The blood vessel system, mostly capillaries, forms rich *reticulum*. The rich nerve *plexus* coming from several smaller nerves runs at the base of the rakers series in the connective tissue of the branchial arch and gives lateral branches to the gill-teeth. The nerves consist of thicker and thinner fibres but structurally they differ from that in the main gill trunk mentioned before. Despite the near position, both of the trunks situated in the connective tissue of the branchial arch no connection could be noted between them.

The nerves of the gill-rakers possess always fewer thick fibres and very many thin ones. They never showed varicosities or coils in their pathways. Very richly branching in the connective tissue they form a dense *plexus* closely connected with the connective tissue cells (Fig. 2). The thick fibres as a rule showed dendronlike ramification or free coils at the end, the thin fibres follow the processes of the connective tissue cells and end freely or with little knobs on the cell bodies or quite next to that.

The small arteries or veins of the connective tissue in the gill teeth seem to have own nerve supply. Small nerve trunks with fine thin nerve fibres or quite

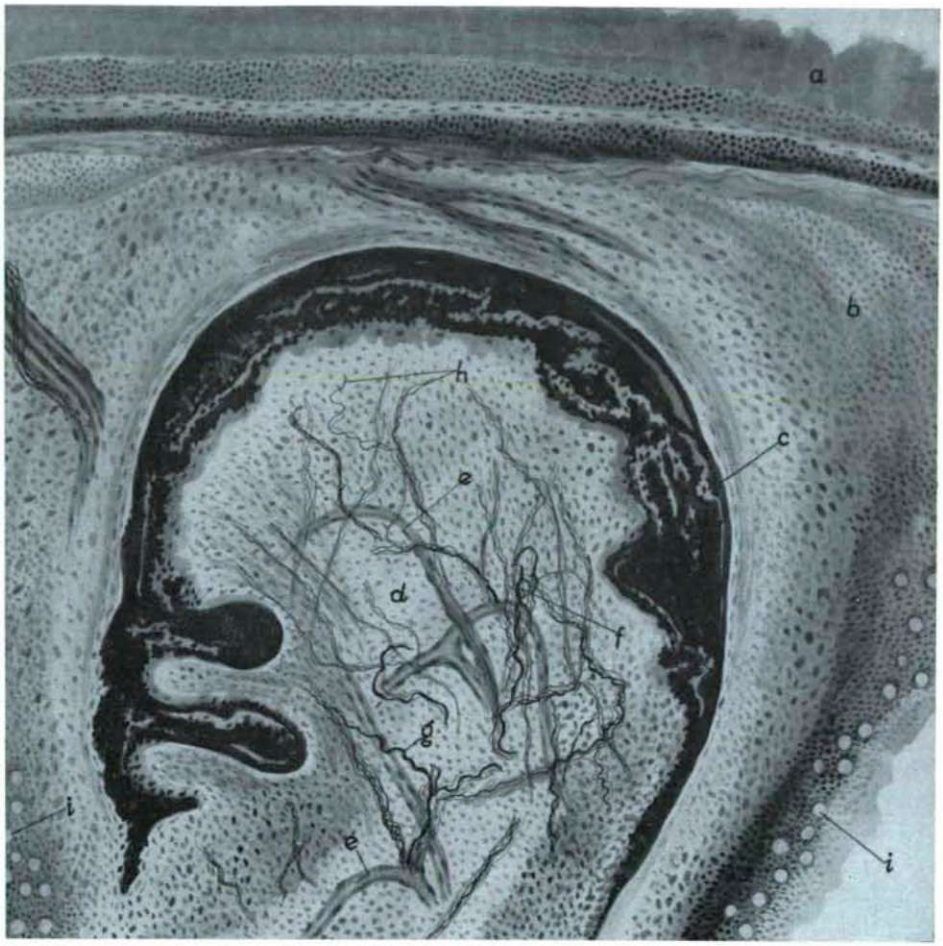


Fig. 2. *Scorpaena porcus:* Structure and innervation of the gill tooth. a — *epithelium* of the mucous membrane, b — *lamina propria,* c —bone rounding the *pulpa,* d — connective tissue of *pulpa,* e — capillaries, f — nerve *plexus,* g — thick nerve fibre, h — thin nerve fibre, i — goblet cell. BIELSCHOWSKY—ÁBRAHÁM's method. Magn. 200x.

separated thin fibres may be connected with the vessel wall. The capillaries are mostly poor of nerve fibres.

The pictures obtained were very similar to the teeth-pulpa innervation of mammals and human. Relatively as many fibres were found in the gill-teeth *pulpa* of *Scorpaena porcus* as by other authors (BERKEBACH VAN DER SPRENKEL 1935, BERNICK 1948, CHRISTENSEN 1938, GORDON-JÖRG 1953, HATTASSY 1958, 1960, HELD- BAUD 1953, LOEWENSTEIN-RATHKAMP 1955, MARTINO 1941) in the teeth *pulpa* of higher vertebrates. So we consider the connective tissue of the gill-teeth as a very sensory area of the gills.

The mucous membrane

In the field of the gill-apparate the whole branchial arches are covered with mucous membrane. The mucous membrane structurally and in respect to its innervation is in full agreement with that of the oral cave. We stated their identity in the *Scorpaena porcus*. The mucous membrane consists of the outer epithelial and the inner connective tissue layers. The *epithelium* is represented by the stratisfied squamous, not keratinized type full of goblet cells. The *lamina propria* shows special structure and forms in places double layer. The higher layer is in close connection with the basal membrane of the *epithelium* and the deeper one is connected with the osseous or muscular elements. Both can be considered as loose connective tissue but in the higher layer the connective tissue cells, in the deeper layer the fibrous elements are dominant. The cell bodies with the processes form *reticulum* in the higher layer, appearing here a very distinct form of the *reticulum* of the connective tissue cells due to the strinking strong affinity to the silver.

The cell *nuclei* have more *nucleoli* and could be observed very frequently their amitotic propagative forms. Numerous small arteries and capillaries were found here and particularly dense nerve *plexus* is characteristic of this layer (Fig. 3). Three kinds of fibres are distinguished in the *plexus*. The three kinds of fibres are located in one trunk in most of the cases. According to their location, structure and end-connections we think the thick fibres (Fig. 3, d) are receptoric fibres of the epithelial cells. The thin ungranulated fibres (Fig. 3, f) must be effectors to the vessels and the glandular (goblet) cells. The fine granulated fibres (Fig. 3, e) which form the greatest part of the fibres, seem to be very sensitive receptoric fibres belonging to the connective tissue cells.

The deeper layer of the lamina propria which may be considered as a *lamina submucosa*, is especially thick in the branchial branch and at the lingual part of the gill-apparate. In this layer, the number of the connective tissue cells are relatively few and the number of the interstitial substance especially that of the collagenous fibres is increased.

This layer surrounds the osseous branchial arch, gives tissue to the *pulpa* of the gill teeth and carries the vessels and nerves to the different parts of the gill-apparate. It is poor of nerves but sometimes free or capsulated coils can be found in this layer (Fig. 4). It is especially interesting because the incapsulated nerve endings are unknown in the fishes, amphibians and reptiles. We could see in the coils thicker and thinner fibres with many longish varicoses in both kinds of fibres. The coils are round or longish and their origin is in all probability from the sensory trigeminal nerve.

The branchial blood vessels

The branchial vessels giving the filamentary branches are lateral branches of the *truncus arteriosus*. They are located in the connective tissue of the branchial arch close to each other the artery, vein and the main nerve trunk of the gill-lamellae. In respect to the innervation of the vessels the artery has always very rich

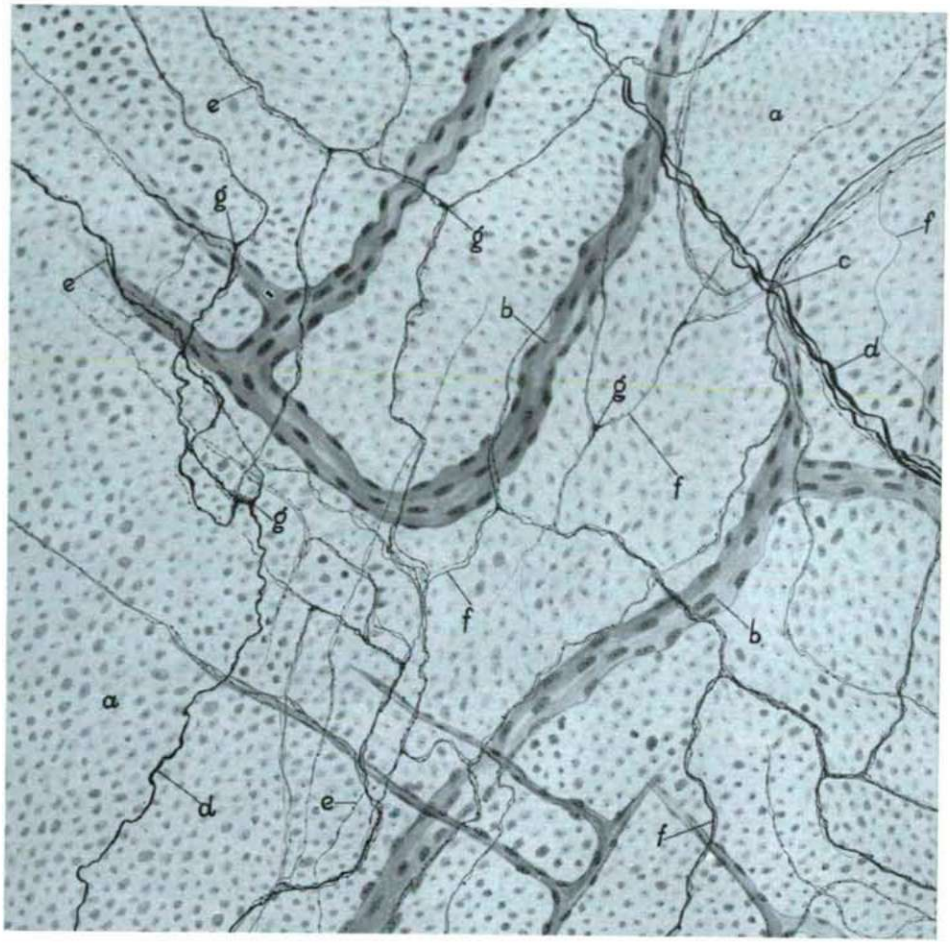


Fig. 3. *Scropaena porcus:* Innervation in the outher layer *of lamina propria* in branchial arch II, a — connective tissue cells, b — capillaries, c — nerve *plexus*, d — thick nerve fibre, e — granulated thin nerve fibre, f — not granulated thin nerve fibre, g — dendron-like branching, BIELSCHOWSKY—ÁBRAHÁM' s method. Magn. 400x.

adventitial *plexus* with very many thick and fewer thinner fibres. The vein is very poor of nerves. In the *plexus* of the branchial artery little groups of nerve cells occurred (Table I. Fig. 4). The cells are multipolar, as a rule 3—6 cells in one group.

Beside the adventitial *plexus* there are some places where special thick nerve fibres were found staining dark black with silver. They are wavy and end with dendron-like forms. (Table II. Fig. 1). Though they have no neurofibrillar endplates at the ends, yet they may be considered as the simpler forms of the *pressoreceptor* system in the arterial trunks of the higher vertebrates. Structurally

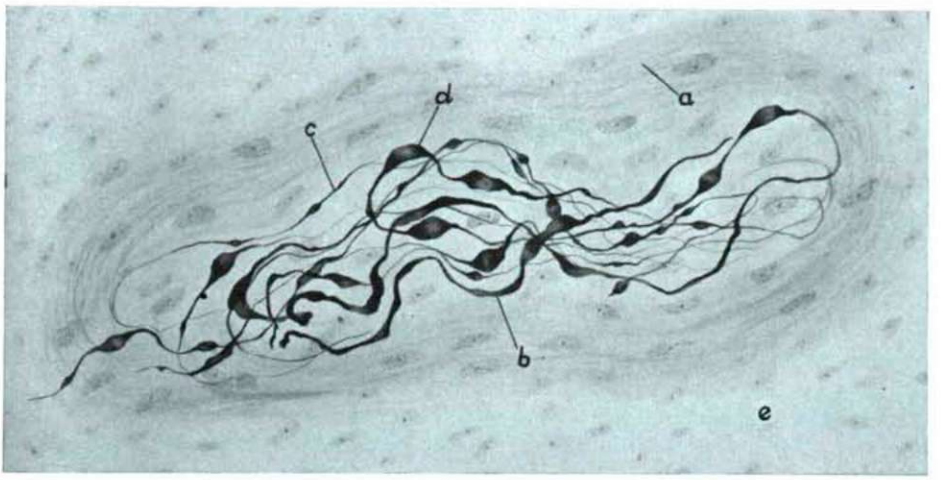


Fig. 4. *Scorpaena porcus:* Capsulated nerve ending in the inner layer of *lamina propria* of branchial arch I, a — capsule, b — thick nerve fibre, c — thin nerve fibre, d — varix, e — interstitial connective tissue. BIELSCHOWSKY—ÁBRAHÁM's method. Magn. 600x.

they are in full agreement with our findings in the swim-bladder (ÁBRAHÁM, STAMMER 1954). Single thick nerve fibres with dendron-like endings appear sometimes on the filamentary arteries, too. They may have similar function, their forms, however, are simpler. We tried to count the number of the pressoreceptors in one gill-apparate and could be stated only 2—4 larger ones in the branchial arteries and 4—6 simpler types in the filamentary arteries. It is true that the I. arch couple has always the largest pressoreceptoric area where 4—6 thick fibres end near each other. Often they were not found in the II. or IV. arches. Where the branchial receptors were absent 3—4 filamentary arteries had receptoric endings.

Quite near the branchial arteries in the connective tissue of the branchial arch some cell groups appeared with intimate nerve connection. Probably they posses similar but simpler structure as the chemoreceptors in higher vertebrates. The nerve trunks coming to these cells contain only thick nerve fibres and show wavy pathway. Their fine end branches are closely connected with the cells (Table II. Fig. 2).

The receptoric end fibres of the branchial vessels found in the *Scorpaena porcus* show the same regulation of the blood circulation in fishes as demonstrated by HEYMANS and NEIL (1960) in the highest vertebrates. Our morphological findings are supported by the physiological data of LUTH-WYMANN, IVNING, SOLANDT, MC. WILLIAM who experienced pressoreceptoric function in *Squalus, Mustelus* and *Anguilla.*

Finally it may be mentioned that the vessels in the gill-teeth and in the mucous membrane were never connected with the branchial vessels and never had any receptoric endings. They are the continuation of the main vessels of the lingual part and the oral cave.

The muscles of the gill apparate

The active movement of the gills in the *Scorpaena porcus* is connected with the well developed branchial-, hyoideal-, and pharyngeal musculature. However, the muscles at the basal part of the filaments, representing the filamentary musculature, seem to be different from the other muscles of the gill apparate. We suppose the differences are in the structure, origin and nerve supply between the two types of the muscles.

The muscle fibres in the filaments are short and thin without any close connection. The muscle bundles run in many directions in the filaments. Their *nuclei* are rounded and rarely visible. Very dense and clear cross striations characterise their myofibrils. The nerve supply of the muscle fibres is very rich. In our opinion on the filamentary muscle fibres get not only one but more nerve end connections. The endings are dendron-like with little end-plates or knobs (Fig. 1, i). The muscle *nuclei*, however, are never collected round the nerve endings. These end-forms are very similar to our findings in the intrinsic musculature of the reptiles and bird's eyes (STAMMER 1962). So we suppose a special innervation of these muscles. It may be that they get not voluntary but autonom motoric innervation.

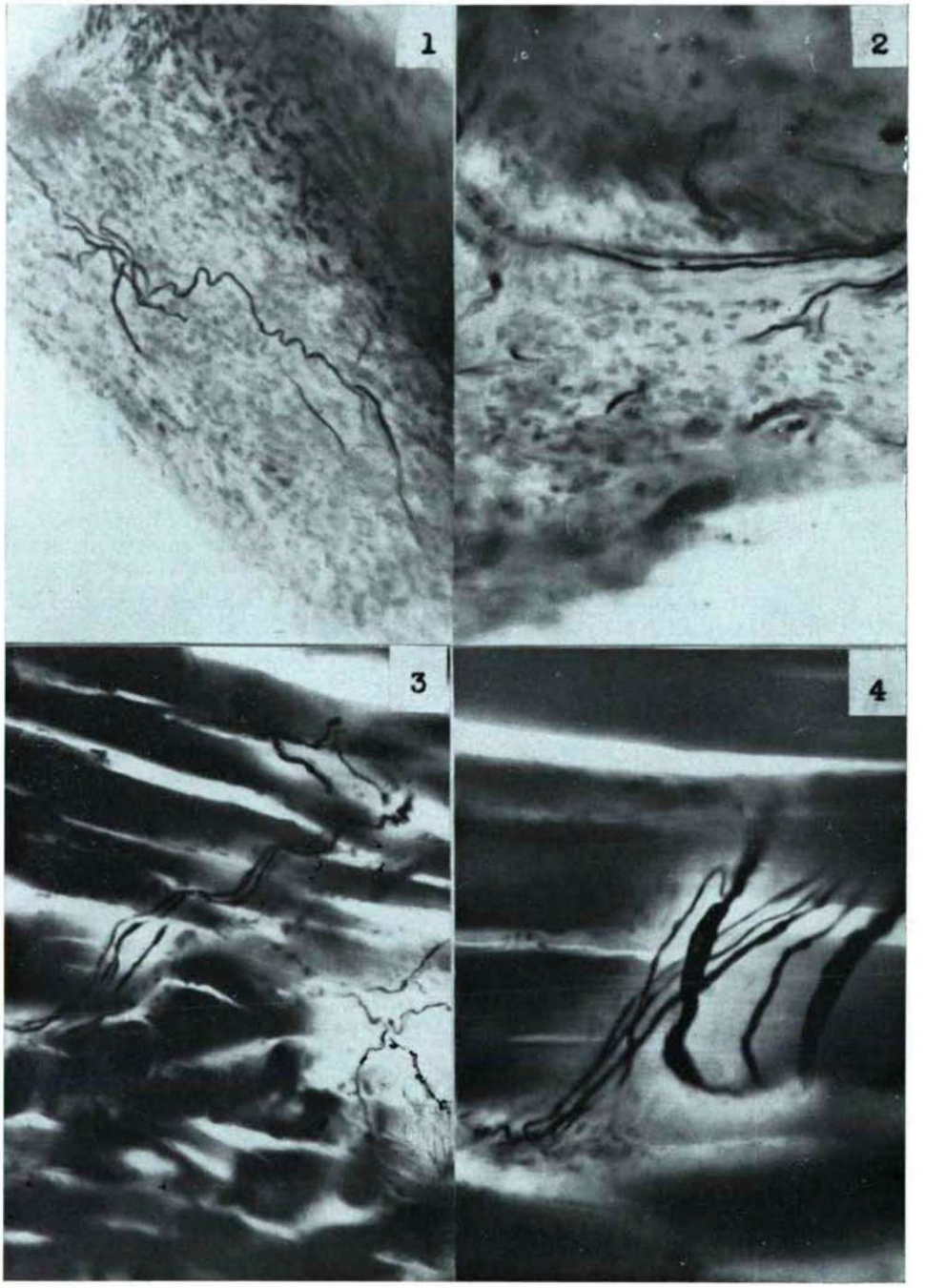
The musculature found at the lingual and pharyngeal ends as well as the groups of the branchial arches, represent the branchial, hyoideal and pharyngeal musculature agree with each other in every respect. These muscles *(Musculus branchialis, hypobranchialis, coracobranchialis, coracohyoideus, pharyngobranchialis* and *mm. interarcuales)* are differentiated to new function from the lateral musculature of the trunk, however, their skeletal characteristics remained. Their fibres showed not so dense and strong cross striation as those mentioned above. The forms of their *nuclei* are always longish and well visible in near position to the *sarcolemma*. Their nerves as everywhere in the skeletal muscles of the fishes consist of very thick and very thin nerve fibres. It is remarkable that the thin fibres appear as lateral side-branches of the thick fibres (Table II. Fig. 3), and the thick fibres become quite thin and after certain distance they are quite thick again. Both appearences were experienced in the skeletal and eye muscles of fresh water fishes (STAMMER 1960). Beside the rapid caliber changes, small or large varices could be noted in the nerve fibres. The nerve trunks of the muscles run obliquely to the parallel muscle fibres, closely connected to each other and give lateral end-branches to both sides (Table II. Fig. 4). The end-forms remind of the motoric end-plates of higher vertebrates but they are simpler and possess poor collection of the muscle *nuclei*.

The nerve trunks run in the muscles segmentally and so there are large areas without any nerve connections. Bearing in mind that the limit of the muscle fibres is roughly at the same level so we consider the segmental innervation where one muscle fibre possesses only one nerve ending.

Table II.

Fig. 1. Pressoreceptoric fibres in the adventitional *plexus* of the branchial artery.
Fig. 2. Chemoreceptoric fibres in the adventitional *plexus* of the branchial artery.
Fig. 3. Segmental innervation of the pharyngeal musculature connected to the gill apparate.
Fig. 4. Motoric nerve ending of the branchial musculature.

TABLE II



The origin of the nerves in the gill apparate

Without experimental examinations is very difficult to speak about this question in fishes. However, to study the degeneration peripherally and centrally which means the only way to solve the problems, seems to be impossible at present. Cutting the nerves or any injury of the gill apparate causes high mortality in fishes.

Considering the obtained pictures of our microscopical examinations, the equalities and differences of the nerve supply as well as the better known nerve connections in highest vertebrates we think about the origin as follows:

1. The common participation of the trigeminal and facial nerves in the mucous membrane.

2. The common participation of the glossopharyngeal and vagal nerves in the branchial area.

3. Sensory fibres of the branchial vessels belonging to the glossopharyngeal and vagal complex.

4. Cranial sympathetic fibres connected always to the plexuses of the vessels, especially to the arteries.

5. The motoric fibres originate from cerebral nerve XII. to the lingual and branchial musculature and from cerebral nerves IX and X. to the pharyngeal musculature.

6. The filamentary muscles get their fibres from the nerve complex of IX. and X. but it may be from the parasympathetic part of these nerves.

7. The gill teeth are innervated only by sensory trigeminal fibres. Their endforms, the sensory coils appeared in the connective tissue of the branchial arch.

* * *

I would express my thanks to Prof. A. Ábrahám (Szeged) for his kind help, Prof. D. Morović (Split) for explaining the life of the sea fishes, and to our assistant Erzsébet Dános for preparing the figures.

Summary

Special structures and innervation was found in the different parts of the gill apparate in *Scorpaena porcus*.

Beside the structurally alternating gill filaments of the gill lamellae, strong gill raker, consist of separated teeth, and well developed musculature are characterised the gill apparate of this animal.

Very rich sensory system was noted in the field of the gill apparate in forms of incapsulated sensory coils and dendron-like end branchings.

The sensory coils are situated in the connective tissue round the branchial arches and inside the gill rakers. The dendron-like end-branchings occurred in the mucous membrane and connected to the branchial vessels.

On the basis of the very rich sensory innervation was stated very important receptoric areas in the gill apparate giving afferent foots to the feeding and breathing reflexes.

The motoric innervation belonged to the musculature and the vessels of the gill apparate.

The author believes the innervation of the gill apparate originates from cerebral nerves V, VII, IX and X and the cranial sympathetic system. They may be abundantly mixed up centrally and peripherally.

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ANGABEN ZUR RHIZOPODEN-FAUNA DER THEISS-STRECKE ZWISCHEN SZOLNOK-CSONGRÁD

D. GÁL

Systematisch-Zoologisches Institut der Attila József Universität Szeged
(Eingegangen am 10. Okt. 1966)

Während der Tisza-Forchungsreise vom 17—30. Juli 1958 hatte lch Gelegenheit, an der Flussstrecke zwischen Tiszapüspöki und Csongrád Sammlungen sowohl aus der „lebenden" Tisza und den in sie mündenden Nebenflüssen (Zagyva und Kőrös), als auch aus den hier liegenden Toten Armen anzustellen. Die Sammlungen erfolgten in Kooperation mit dr. Margarete Szabados und Zoltán Jósa. In der vorliegenden Arbeit sollen nur die Rhizopoden des fliessenden Wassers mitgeteilt werden.

Untersuchungsmethoden

Aus den Flüssen wurde hauptsächlich Plankton an verschiedenen Stellen und ausserdem hie und da auch Schabsel von im Wasser liegenden Steinen und anderen Gegenständen gesammelt. Das Material wurde grösstenteils in offenen Gläsern lebend aufbewahrt und kleinere Portionen auch mit Formalin bzw. Sublimat fixiert. Leider konnte ich das fixierte Sammelgut nicht verwerten.

Gleich nach meiner Heimkehr nahm ich eine Musterung des gesamten Materials vor, registrierte die gefundenen Arten um feststellen zu können, ob während der Aufbewahrung die Artenzusammensetzung keine Veränderung erleidet. Die eingehende Aufarbeitung und Bestimmung erfolgte erst später. Bei der Bestimmung fand ich, dass die Artenzusammensetzung nur zu einem geringen Prozentsatz verändert war, die Individuenzahl sich aber vermehrt hatte.

Bei der Aufarbeitung betrachtete ich jede Phiole als ein besonderes Biotop und untersuchte, was für Arten an der Oberfläche, in der mittleren Wasserschicht und in dem sedimentierten Detritus leben. Die Untersuchungen wurden in erster Linie an lebenden Tieren vorgenommen, nach entsprechender Beobachtungszeit fixierte ich aber auch Material in Essigsäure-Formol-Sublimat und konnte so in den meisten Fällen auch den Kern sichtbar machen.

Die Rhizopoden der einzelnen Sammelstellen

1. In Höhe von Tiszapüspöki (346. Flusskilometer) Plankton aus der Theiss: Oberfläche arm an Tieren, lediglich einige *Arcella rotunda* var. *aplanata* kommen zum Vorschein. (GROSPIETSCH (1958) nach lebt sie in Subsmersmoos). Als viel bevölkerter erweist sich der Detritus sowohl hinsichtlich der Ar-

ten- als auch der Individuenzahl. Dominierende Arten sind *Arcella rotunda* var. *aplanata* und *Trinema lineare*. Ihre Individuenzahl übertrifft die aller übrigen Arten bei weitem. In relativ grosser Zahl findet sich eine kleinere Form (20—22 μ) der *Amoeba limax*, die sich sehr schnell bewegt. Die übrigen Arten sind in niedriger Individuenzahl vertreten: *Amoeba gorgonia, Centropyxis aculeata, C. contstricta, Arcella gibbosa, A. hemisphaerica, Difflugia gramen, Euglypha brachiata, Cryptodifflugia oviformis.* Die angeführten Arten kamen in annähernd gleicher Individuenzahl vor, ausgenommen *Euglypha brachiata*, von der ich nur ein Exemplar vorfand.

2. Beim Kovácsi Fok (340. Fluss-km) Plankton aus der Theiss. Bei der ersten übersichtlichen Untersuchung fand ich an der Oberfläche reichlich *Vahlkampfia debilis*, die bei der späteren eingehenden Untersuchung nicht mehr gesichtet wurden. Hieraus darf es folgen, dass *Vahlkampfia debilis* eine ausgesprochene Flusswasserart ist (auch HARNISCH [1960] nach lebt sie in Flusswasser). Daneben fanden sich an der Oberfläche noch einige *Arcella rotunda* var. *aplanata*. Es waren im Detritus *Arcella rotunda* var. *aplanata, Trinema lineare* und *Euglypha alveolata* in sehr hoher Individuenzahl, nebenbei kamen auch einige *Pareuglypha reticulata* und *Difflugia gramen* vor.

3. *Ceratophyllum* und Holzrindenstück vom Wasserrande beim Kovácsi Fok: bei der ersten Untersuchung sah ich auch hier *Vahlkampfia debilis*, die aber später verschwunden war. Unter den Pflanzen dominieren die Testaceen. In grösserer Zahl kamen *Trinema enchelys* und in geringerer *Trinema lineare* vor. Von den Schalenlosen herrscht *Hyalodiscus korotnevi* vor, in geringerer Individuenzahl findet sich *Amoeba gorgonia*.

4. Schlamm aus der Theiss beim Kovácsi Fok: im Schlamm sehr zahlreich *Amoeba limicola* und einige *Arcella discoides*.

5. Schabsel von am Ufer liegenden Steinen beim Kovácsi Fok: Rhizopoden wurden nicht gefunden.

6. Schaumiger Belag von der Wasseroberfläche am rechten Tiszaufer bei Szolnok, etwa 200 m von der Brücke entfernt: sehr arm an Rhizopodenarten, lediglich einige Exemplare von *Arcella rotunda* war. *aplanata* und *Trinema lineare*.

7. Plankton aus der Theiss bei der 6. Sammelstelle: an der Oberfläche leben ausschliesslich schalenlose Rhizopoden. Mit sehr hoher Individuenzahl dominiert *Vahlkampfia mira* (ausgewiesen von HARNISCH [1960] im Schlamm des Sees) daneben sind in geringer Individuenzahl *Amoeba guttula* auffindbar. Im Detrius dominieren *Trinema lineare* und *Cyphoderia laevis* in grosser Individuenzahl. Daneben gelangen einige Exemplare der schon erwähnten kleineren Variante von *Amoeba limax* zur Beobachtung.

8. Plankton aus der Zagyva 200 m von der Brükke aufwärts: Im Detritus herrschen *Arcella rotunda* var. *aplanata* und *Cyphoderia margaritacea* von, während *Trinema lineare* in etwas geringerer Individuenzahl vertreten ist. Ausserdem kamen 4 *Arcella vulgaris*-Exemplare und auch 1 stück *Difflugia gramen* zur Beobachtung. Eine für die Zagyva typische Testaceenart kommt in grosser Individuenzahl vor, leider konnte ich lebende Exemplare nicht entdecken, sondern nur leere Schalen. Die Schalen sind seitlich abgeplattet und an der Oberfläche mit Quarzkörnchen bedeckt. Der Mund ist endständig, rund und von

Quarkörnchen umgeben. Die Schale hat bräunliche Farbe, ihr hinteres Ende verlauft spitz. Länge: 105. μ, Breite: 60 bzw. 70 μ. (Tafel II. 9 a und b).

9 .Plankton aus der Zagyva, 15 m von der Brücke abwärts, beim Abwasserabfluss: An der Oberfläche ist das massenhafte Erscheinen von *Amoeba beryllifera* charakteristisch, oft kommen 10—15 Exemplare in ein Gesichtsfeld. Dominierende Arten des Detritus sind *Arcella rotunda* var. *aplanata* und *Trinema lineare*. In hoher Individuenzahl finden sich noch *Cyphoderia margaritacea*. Von den schalenlosen Rhizopoden fand ich nur 1 einziges Exemplar der *Amoeba vespertilio* vor. In relativ grosser Individuenzahl kamen auch die in dem vorhengehenden Material geschilderten Testaceenarten vor.

10. Plankton aus der Theiss in einer Einbuchtung des linken Ufers beim 327. Kilometerstein: sowohl an der Oberfläche, als auch im Detritus leben nur reichliche *Arcella rotunda* var. *aplanata*, im Detritus in etwas grösserer Individuenzahl.

11. Dortselbst, Belag eines im Wasser liegenden Borkenstückes: Das Schabsel enthält reichlich *Trinema lineare* und *Euglypha laevis*. Daneben finden sich *Euglypha ciliata* und *Cryptodifflugia oviformis* in grösserer Zahl. *Amoeba terricola* war mit 2 Exemplaren *Centropyxis constricta* und *Euglypha brachiata* mit je einen Exemplar vertreten.

12. Dortselbst, morsches Holz im Wasser: Dominierende Art ist *Centropyxis aculeata*, die mit zahlreichen Individuen vertreten ist. Von *Centropyxis constricta* wurden weit weniger Exemplare gesichtet und *Trinema lineare* und *Pareuglypha reticulata* kommen nur in sehr kleiner Individuenzahl vor.

13. Dortselbst, aus einer sickernden Vertiefung auf Löszboden geschöpftes Wasser: Rhizopoden kommen weder bei der ersten, noch bei der späteren, eingehenden Untersuchung zum Vorschein.

14. Dortselbst, aus einer Vertiefung zwischen Uferabgrung und Wasser: an der Oberfläche reichlich *Vahlkampfia mira* und einige Exemplare einer *Pelomyxa*-artigen Rhizopode.

15. Plankton vom rechten Theissufer bei Vezseny: auffallend reich an Rhizopoden. An der Oberfläche zahlreiche *Cochliopodium obscurum* (*Gocevia obscurum* PENARD; nach HARNISCH [1960] sind sie an Seenfern und kleinen Gewässern zu fiden), daneben einige *Amoeba gorgonia*. Auch von *Centropyxis constricta*, *Arcella rotunda* var. *aplanata* und *Arcella gibossa* war je 1 Exemplar anwesend. Ausserdem fand ich je 1 Exemplar zweier bisher nicht bestimmter Amoebenarten (ihre Beschreibung erfolgt am Ende dieses Kapitels [*Amoeba* sp. I., II.]). Im Detritus erschienen *Trinema lineare*-Individuen in riesigen Mengen. In geringerer Individuenzahl fand ich *Arcella rotunda* var. *aplanata* und *Arcella gibbosa*, nebenbei auch einige Centropyxis constricta.

16. Plankton aus der Theiss am linken Ufer bei Vezseny: trotz des Bakterienreichtums an der Oberfläche, die sonst eine gute Nahrung für die Amoeben darstellen, fand ich nur 2 *Arcella rotunda* var. *aplanata*. Im Detritus erschienen *Cyphoderia laevis* massenhaft in geringerer Zahl *Trinema lineare* und *Arcella rotunda* var. *aplanata*, sowie einige *Arcella gibbosa*, *Centropyxis constricta* und *Pareuglypha reticulata*.

17. Schabsel von der Seite eines Schleppschiffes oberhalb des Wasserniveaus: enthält keine Rhizopodenarten.

18. Schabsel von der Seite eines Schleppschiffes in Höhe des Wasserniveaus: Ich fand eine einzige Rhizopodenart vor, deren Bestimmung mir bisher nicht gelungen ist: häufig ist die ganze Körperoberfläche mit dünnen, zugespitzten Pseudopodien bedeckt, die wasserklar und druchsichtig sind. Entoplasma wenig granuliert, darin werden einige grosse rote und blaue, verschiedenförmige Granulae sichtbar und auch eine verhältnismässig grosse Vakuole. Der Körper ist oft sphärisch rund und ringsum von zugespitzten Pseudopodien umgeben. Der Körperdurchmesser beträgt dann rund 40 μ, gestreckt 75—80 μ (*Amoeba* sp. III. Tafel I. 3. a und b).

19. Schabsel von einem Schleppschiff unterhalb des Wasserspiegels: die gleiche *Amoeba* sp. III. wie an der Wasseroberfläche.

20. Plankton von der Flussmitte bei Tiszaug: an der Oberfläche nur zahlreiche *Arcella rotunda* var. *aplanata*, die den Detritus überaus reich bevölkern. Daneben kamen einige *Arcella gibbosa* (von GROSPIETSCH [1958] in sauren Gewässern angezeigt) und *Centropyxis constricta* zum Vorschein.

21. Detritus und Algenüberzug von Steinen am linken Theissufer bei Tiszaug: hier dominiert ebenfalls *Arcella rotunda* var. *aplanata* in gewaltigen Mengen. In fast ebenso grossen Mengen finden sich auch *Pareuglypha reticulata* und etwas weniger reichlich *Pyxidicula operculata*. Auch einige *Arcella gibbosa*, *Euglypha elveolata* und *Euglypha brachiata* sind vorhanden.

22. Plankton vom rechten Ufer bei Tiszaug: an der Oberfläche sehr viele *Arcella rotunda* var. *aplanata* und 2 *Amoeba vespertilio*-Exemplare. Dominierende Arten im Detritus sind *Arcella rotunda* var. *aplanata*, *Trinema lineare* und *Pareuglypha reticulata*, danaben leben *Cyphoderia laevis* und *Euglypha laevis*.

23. Plankton aus der Theiss bei Tiszaug etwa 200 m von der Brücke, vor der Einmündung des Toten Armes: an der Oberfläche neben zahlreichen *Amoeba limicola* 1 Exemplar von *Arcella rotunda* var. *aplanata*. Im Detritus ausser zahlreichen *Cyphoderia laevis* und *Trinema lineare* wenige *Arcella rotunda* var. *aplanata*.

24. Planktonentnahme unter einem Schiff bei Tiszaug: an der Oberfläche nur einzelne *Arcella rotunda* var. *aplanata*. Im Detritus dominieren *Arcella rotunda* var. *aplanata* und *Trinema lineare*. In geringer Individuenzahl wurden *Arcella vulgaris*, *Arcella gibbosa*, *Centropyxis constricta* und *Cyphoderia laevis* gesichtet.

25. Plankton aus der Kőrös — Mündung am rechten Ufer: an der Oberfläche zahlreiche Bakterien, aber keine Rhizopoden und auch im Detritus nur einige *Trinema lineare*.

26. Plankton aus steinumgebenen Buchten am linken Kőrösufer zwischen Kunszentmárton und Szelevény: an der Oberfläche *Amoeba limicola* in grosser Zahl. Im Detritus dominiert *Trinema lineare*, daneben finden sich reichlich *Amoeba limicola* und *Pareuglypha reticulata*, sowie einige *Euglypha brachiata*, *Centropyxis constricta*, *Euglypha alveolata*, *Difflugia mammillaris*, *Pseudodifflugia fascicularis* und *Trinema enchelys*.

Artenbeschreibungen

1. *Amoeba* sp. I.
(Tafel I. 1.)

Sie kam aus dem bei V e z s e n y am rechten Tiszaufer entnommenen Plankton zum Vorschein. Körperform vieleckig, in den meisten Fällen 3 — bzw. 4-eckig, an der Polen im Verhältnis zum Körper sehr lange, überaus feine, biegsame, schnellbewegliche Pseudopodien. Seltener bilden sich an der einen Seite einige kürzere, dickere, fingerartige Pseudopodien. Das Entoplasma ist relativ kompakt, granuliert und ohne Ektoplasmasaum. In der Regel eine grosse Vakuole, aber es wurden bisweilen auch vier kleine Vakuolen sichtbar. Körperdurchmesser 8 μ, maximale Pseudopodienlänge 20—22 μ.

2. *Amoeba* sp. II.
(Tafel I. 2. a und b).

Sie kam zusammenen mit *Amoeba* sp. I. zur Beobachtung. Morphologisch waren zwei Typen zu unterscheiden: die häufigere Form bildet an der Hinterseite 1—5 lange, dicke, zugespitzte Pseudopodien, während der Vorderteil abgerundet und meistens ohne Ektoplasmasaum ist. Die Entoplasmakörnchen dringen auch in den Initialabschnitt der Pseudopodien vor. Die andere Form geht aus dieser hervor, indem sich vorne ein breiter Ektoplasmasaum bildet und hinten die langen Pseudopodien „resorbiert" werden. Sehr lange besteht sie in einer Form, wo hinten nur ein dickes Pseudopodium vorliegt, und ist dann sehr schnell beweglich. Eine grosse Vakuole. Körperdurchmesser 38—40 μ, die Pseudopodien können eine Länge bis zu 60—70 μ erreichen.

3. *Pelomyxa* sp.

Meistens ellipsoide Form, langsam beweglich. Bildet keine Pseudopodien, nur selten befindet sich vorn ein sehr breiter, kurzer Plasmawulst. Am Vorderteil ein schmaler, wasserklarer Ektoplasmassaum. Das Entoplasma ist granuliert, darin im Vorderteil 8—10 Vakuolen, von denen eine wächst grösser an und pulsiert schneller, als die übrigen. Länge 35 μ.

Zusammenfassung

Wie es aus den Tabellen I. und II. erhellt, leben and der Oberfläche des Sammelmaterials nur sehr wenig Rhizopodenarten, ungeachtet dessen, dass das gesammelte Material grosstenteils aus dem Oberflächenplankton stammt. Dies lässt vermuten, dass für den grössten Teil der Rhizopoden-Arten — in erster Linie für die Testaceen — nicht das Plankton das optimale Biotop darstellt und diese lieber im Schlamm leben. Ihr Vorkommen im Plankton ist nur eine sekundäre Erscheinung; die Wasserströmung treibt sie aus dem Schlamm aufwärts und so gelangen sie ins Plankton.

In der T h e i s s kommen in erster Linie die Testaceen in grösserer Arten — und Individuenzahl vor. Dominante Arten sind *Arcella rotunda* var. *aplanata*

und *Trinema lineare*. *Arcella rotunda* var *aplanata* scheint sich auch in der Oberflächenmembran wohl zu fühlen, wo sie von den Rhizopodenarten die häufigste ist. Von den Testaceen kam an der Oberfläche ausser der *Arcella rotunda* var. *aplanata* je einmal auch *Arcella gibbosa*, *Centropyxis constricta* und *Trinema lineare* — aber alle nur in geringer Individuenzahl — vor. Unter den Amoeben habe ich allgemein verbreitete Arten nicht gefunden, sie kamen höchstens an 1 oder 2 Sammelstellen vor.

Die beiden aus der Zagyva entnommenen Planktonprobem weichen stark von dem Material der Theiss hinsichtlich des massenhaften Vorkommens der beschriebenen Testaceenart und der *Cyphoderia margaritacea* ab. An der Oberfläche der nahe des Abwasserausflusses entnommenen Planktonproben lebt *Amoeba beryllifera*, die ich in der Theiss nicht auffand.

In der Kőrös sind es *Amoeba alveolata*, *Penardia cometa*, *Difflugia mammillaris* und *Pseudodifflugia fascicularis*, die in der Theiss nicht nachweisbar sind.

Die bisher gefundenen Arten ergeben sich zusammenfassend:

aus der Theiss 32 Arten

aus der Zagyva 8 Arten (3 davon habe ich in der Theiss bisher nicht gefunden)

aus der Kőrös 11 Arten (4 davon kamen bisher in der Theiss nicht zur Beobachtung).

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Tabelle I. An der Oberfläche gefundene Arten

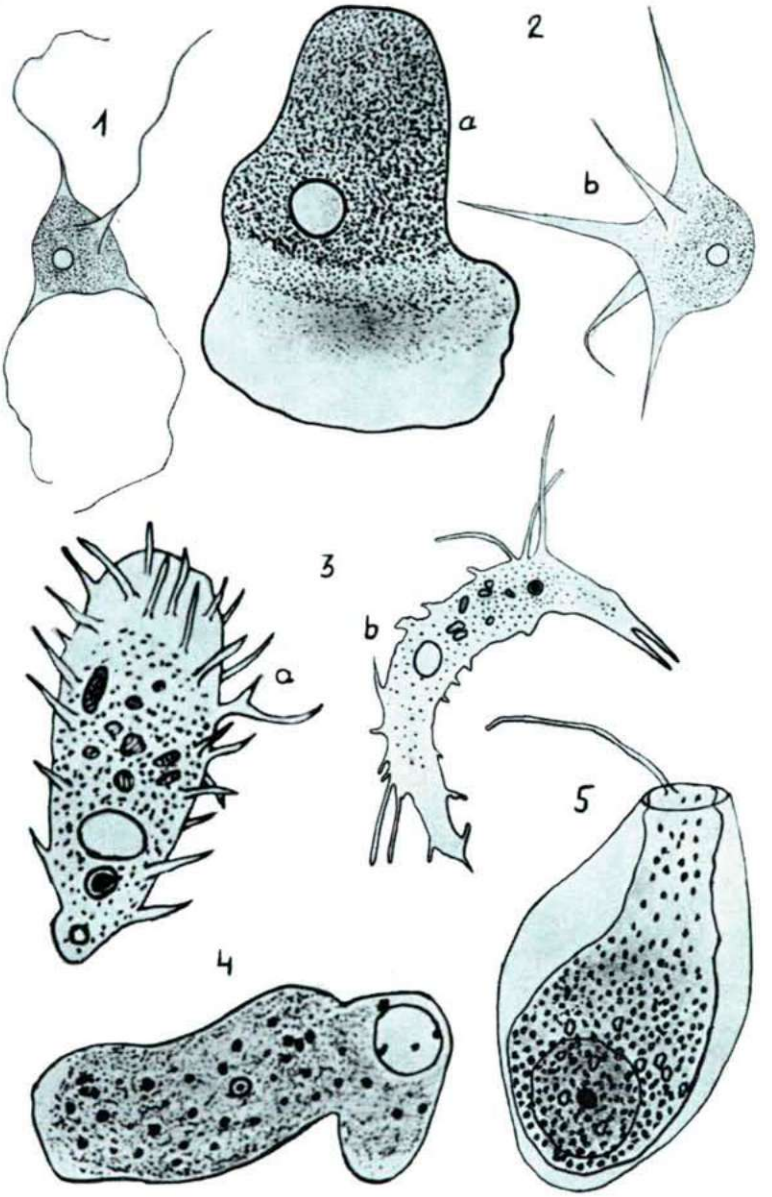
	1	2	3	6	7	9	10	14	15	16	18	19	20	22	23	24	26	27
Amoeba guttula DUJ.					2													
Amoeba limicola RHUMBLER															4		4	
Amoeba beryllifera PENARD						4												
Amoeba gorgonia PENARD									2									
Amoeba vespertilio PENARD														1				
Amoeba alveolata MERESCHKOVSKY																		2
Amoeba sp. I.									1									
Amoeba sp. II.									1									
Amoeba sp. III.																		
Vahlkampfia debilis JOLLOS		4	4								3	3						
Vahlkampfia mira GLÄSER					4			4										
Pelomyxa sp.								2										
Penardia cometa (PEN.) SAEDELEER																		2
Cochliopodium obscurum PENARD									4									
Centropyxis constricta DEFL.									2									
Arcella gibbosa PENARD									2									
A. rotunda var. aplanata DEFL.	2	2		2			3		2	1			4	4	1	3		
Trinema lineare PENARD				2														

Tabelle II. Im Detritus gefundene Arten

	1	2	3	4	7	8	9	10	11	12	15	16	18	19	20	21	22	23	24	25	26	27
Amoeba limax DUJARDIN	3				2																	
Amoeba limicola RHUMBLER				4																	3	
Amoeba gorgonia PENARD	2		2																			
Amoeba vespertilio PENARD							1															
Amoeba alveolata MERESCHKOVSKY																						4
Amoeba terricola EHRBG.									1													
Amoeba sp. III.													3	3								
Hyalodiscus korotnevi MERESCHKOVSKY			3																			
Difflugia mammillaris PENARD																					2	
Difflugia gramen PENARD	2	2				1																
Centropyxis aculeata STEIN	2									4												
Centropyxis constricta DEFL.	2								1	3	2	2			2				2		2	4
Arcella vulgaris EHRBG.						2													2			
Arcella discoides EHRBG.				2																		
Arcella gibbosa PENARD	2										3	2			2	2						
A. rotunda var. aplanata DEFL.	4	4				4	4	4			3	3			4	4	4	2	4			
Arcella hemisphaerica PERTY	2																					
Pyxidicula operculata EHRBG.																3						
Cryptodifflugia oviformis PENARD	2								3													
Pseudodifflugia fascicularis PENARD																					1	
Cyphoderia laevis PENARD					4							4					2	4	2			
Cyphoderia margaritacea EHRBG.						4	3															
Pareuglypha reticulata PENARD		2								2		2				4	4				3	
Euglypha alveolata LEIDY		4														2					2	2
Euglypha ciliata EHRBG.									3													
Euglypha brachiata LEIDY	1								1							2					2	
Euglypha laevis PERTY									4								2					
Trinema lineare PENARD	4	4	3		4	3	4		4	2	4	3					4	4	4	2	4	2
Trinema enchelys EHRBG.			4																			
Testacea sp.						4	3															

Zeichenerklärung zu Tabellen I. und II.: 4 = in grosser, 3 = in mittlerer und 2 = in geringer Individuenzahl erscheinene Arten, 1 = 1—2 Individuen.

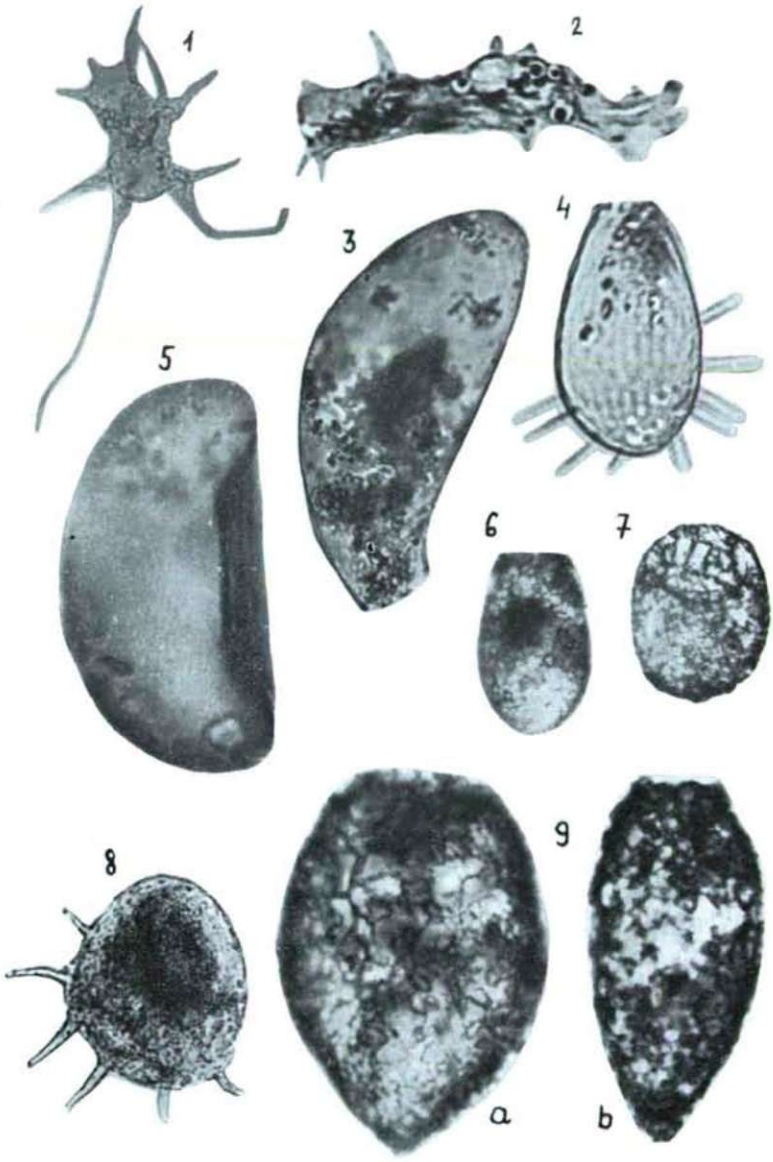
TAFEL I.



Tafelerklärung zu Tafel I.:

1. *Amoeba* sp. I.
2. a. b. *Amoeba* sp. II.
3. a. b. *Amoeba* sp. III.
4. *Amoeba beryllifera*
5. *Cyphoderia laevis*

TAFEL II.



Tafelerklärung zu Tafel II.:

1. *Amoeba vespertilio*
2. *Amoeba alveolata*
3. *Cyphoderia margaritacea*
4. *Euglypha ciliata*
5. *Arcella rotunda* var. *aplanata*
6. *Difflugia gramen*
7. *Centropyxis constricta*
8. *Centropyxis aculeata*
9. a. b. *Testacea* sp.

ÜBER TRIASKORALLENFAUNA UNGARNS.

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(Eingegangen am 29. Sept. 1966)

In: „Enumeration des coraux triassiques de la Hongrie" (Bull. Mens. Soc. Linnéenne de Lyon 1956, 25 année, No. 9. p. 231—232) hatte ich eine provisorische Liste über die Triaskorallen aus dem Gebiete der Ungarischen Volksrepublik gegeben. Die andere Liste von K. PAPP (13) über die Triaskorallen des Bakonygebirges bei Veszprém ist auch schon veraltet aber wegen Mangel der Objekte beider Materialien wurden hier nur diejenige Objekte revidiert und neubearbeitet, welche für Untersuchung mit in meine Szegediner Universität-Sammlung zur Verfügung standen. — Dieses Material gehört zur Zeit dem Institute für Zoosystematik der Universität Szeged und wurde in den letzten Jahrzehnten von FR. LEGÁNYI, K. BALOGH, Frau VÉGH, A. KASZAP, G. CSIZMAZIA und vom Verfasser aufgesammelt. Die, in meinem oben erwähnten französischen Artikel und die in der PAPP'schen Liste (13) bekanntgegebenen Korallenarten der Trias sind hier nicht alle revidiert und neubearbeitet, weder nomenklatorisch noch in merito — so, dass man feststellen muss, dass meine hiesige Besprechungen der Triaskorallenarten nicht auf alle mögliche und bisher von mir (6—10) und von PAPP (13) publizierte Artenangaben sich beziehen. Während meiner Revision und Neubearbeitungen wurden einige neue (d. h. von mir bisher nicht publizierte) Entdeckungen ermöglicht, so, dass die hiesige Publikation mit einem kleinen, aber wichtigen Vorschritt in der Kenntnis unserer heimischen Fossil-Faunistik wertbar ist. Es wurden so: *Plerophyllum, Triadophyllum* usw... zum ersten male auch als Arten betrachtet bezw. beschrieben, sowie andere (vorher nicht genau und richtig bestimmte) Arten synomymisiert oder nullifiziert worden.

Hier finden wir Angaben aus der Mecsekgebirge, Bükkgebirge, Budaergebirge, Keszthelyergebirge und Cserhátergebirge der Ungarischen Volksrepublik. Natürlich sind meine Angaben — die in Bull. Mens. Soc. Linnéenne de Lyon 1956 publiziert waren nicht alle unrecht, so, dass die meisten Angaben für unsere Mittelgebirgen bilden auch jetzt eine gute Ausgangspunkt für weitere Untersuchungen in der Zukunft.

Plerophyllum cf. cuneatum ILJINA

Taf. II. Fig. 2.

Fundort: Bükkgebirge bei Felsőtárkány. In graue Kalke eingebettet. Ein Polypenfragment geschlifft. Durchmesser cca 20 mm Epithek verloren. Der Fund scheint fast vollkommen übereinstimmend mit der Fund ILJINAS (1) und s. auch ihre Tab. X. Fig. 2a. — Nach ILJINA (1) ist die Art eine „nizsnij"-triassische. Unser Fund ist der erste aus der Trias von Ungarn. In der permischen Schichten der Bükkgebirge wurden schon längst durch meine Arbeiterungen mehrere Plerophyllen beschrieben — welche aber nicht mit der hier besprochene Art identisch erscheinen.

Triadophyllum cf. posthumun WEISSERMEL

Taf. II. Fig. 7. und 8.

Fundort: Bükkgebirge — anisische Schichten, in dunkelgrauen Kalke eingebettet. Mehrere Polypen, in verschiedenen Lebensalter bewahrt. Sie scheinen gruppenweise vorzukommen. Vergleichen wir nun unsere Funde mit den weissermelschen Angaben aus der deutschen Muschelkalke (14, 15) so kommen wir zu den folgenden Resultat, welches für uns eine Identitätswahrscheinlichkeit zeigen erscheint:

Eigenschaften Funde	Polyp	Kelch mm	Kelch Tiefe	Theka	Septenanzahl	Septen in Zentr.	Vorkommen
Bükkgebirge	Cylin. Trichter	20	tief	dick	60—62	28	Grupp.
Deutsche Muschelkalk	Trichter	17—18	13,5 mm	kräftig	32—66	33	Solo

Montlivaltia sp. indet

Mehrere Überreste von Bükkgebirge in weissem mitteltriassischen Kalke, sowie im Recoaro-Kalke des Misinagipfels in Mecsekgebirge. Die letztere Funde scheinen einen kleinen Wahrscheinlichkeit zu die Art *Montlivaltia cf. solomoni* WEISSERMEL zu ziehen (14, 15) s. unser Photo auf Taf. II. Fig. 3.

Montlivaltia obliqua (MÜNSTER)

Fundorten: Bükkgebirge unter B á n y a-Berg und H á m o r in weissem mitteltriassischen Kalke eingebettet. Eine gemeine Art der Mitteltrias der Alpenländer, Karpatenländer und Pannonien.

Montlivaltia legányii n. sp.

Fundorten: Bükkgebirge in B a l l a-Gipfel und neben dem Gipfel, zusammen zwei Exemplare in weissen Kalke eingebettet. Beide sind Riesen-Polypen mit Kelchdurchmesser von 60x100 und 90x45 mm. Kelch oval. Sehr ähnlich zu der Art der Oberen Trias *Montlivaltia marmorea* FRECH welche aber einen ganz runden kreisartigen Kelchkontur besitzt. Die Septenanzahl unserer Polypen ist über 200; Endothek sehr dicht und fein, Wand dünn, Septen lang und fein gebogen, und in allen Ordnungen gleich entwickelt. Das Kelchzentrum d. h. *Proto*-oder *Monostomium* länglich-linear. Die Verlängerung ist lateralseits ausgebildet.

Monilivaltia norica FRECH, SQUIRES, KOLOSVÁRY

Fundorten: Bakonygebirge bei Nagycsákány und Csákánypuszta; Bükkgebirge bei Répáshuta; Keszthelyergebirge bei Vállus und Budaergebirge bei Lipótmező und Remete-Berg.

Weit verbreitete, gemeine, stark variierende, sehr elastische Art der Mittel- und Oberen Trias, gemein in Karpatenländer, in Alpenländer und Pannonien. Über eine Identifikation und Kritik s. näheres über frechschen Typ in meinen Publikationen I—V über die Triaskorallen der C. S. S. R. in „*Geologické Práce*" 1956—1965!

Thecosmilia difilippi STOPPANI

Fundorten: Bakonygebirge bei Csengőhegy und Alsóperepuszta, sowie bei Dudar in obertriassischem Kalke eingebettet. Eine Koloniebildende Koralle mit akmischen grossen Polypendurchmesser von 12—15 mm und mit parakmischen lockeren Polypenröhrenstellungen innerhalb der Kolonie. Das ist schon eine Desorganisationserscheinung betreffs der Phylogenie. Polypenröhren können miteinenader mit Kommissuren zusammenhängen.

Thecosmilia cespitosa REUSS

Taf. I. Fig. 6. und 7.

Fundorten: Budaergebirge bei Nagykovácsi und in Triaslinse von Lábatlan. Kelch tief. Polypenröhrenstellung originell dicht, epistatisch. Septen starr und schlank entwickelt. Zentrum des Kelches länglich-oval. Nur obertriassisch.

Thecosmilia badiotica VOLZ

Fundorten: Bükkgebirge unter Bánya-Berg, Répáshuta, Felsőtárkány, Subalyuk, sowie Bükkszentkereszt (Tannenallee) und in Bükk-Plateau — überall in weissem mitteltriassischen Kalke: kalk- und bankbildend. Cserhátergebrige Csővár.

Die Art ist sehr gemein in Mitteltrias der Alpenländer, Karpatenländer und Pannonien. Die Kolonien sind stellenweise in verschiedenen Grade der Umwandlung festzustellen als ein Beweis dafür, dass es organogene d. h. koralligene Kalkbildung recht vorkommt. Polypendurchmesser bis 7 mm. Polypenröhrenstellung bischen locker.

Thecosmilia subdichotoma (MÜNSTER)

Taf. III. Fig. 1.

Fundorten: Bükkgebirge bei Zsércnagydél und Bükkszentkereszt samt *Thecosmilia badiotica* bank- und kalkbildend. Polypendurchmesser nur 5 mm. Polypenröhrenstellung dicht. Epistatisch. Im Kalke in ver-

schiedenen Umwadlungsstadien vorkommend. Auch ein Beweis dafür, dass es organogene d. h. *Thecosmilia*-Kalke gibt. Gemeine Art der Mitteltrias der Alpen-Karpaten und Pannonischen Länder.

Thecosmilia granulata (KLIPSTEIN)

Taf. III. Fig. 3.

Fundorten: Bükkgebirge unter B á n y a-Berg, R é p á s h u t a und B ü k k s z e n t k e r e s z t am Wald-Rand-Weg. Eine Art der Mitteltrias. Polypen winzig, 3—4 Individuen in eine Gruppe vergesellschaftet. Typisch epistatisch.

Thecosmilia clathrata (EMMRICH)

Fundorten: Budaergebirge bei R e m e t e-Berg und M á r i a r e m e t e, sowie in Pannonien bei S ü m e g am Tapolcaer Weg. Auch in Cserhátergebirge bei C s ő v á r gefunden. Polyperöhrenstellung epistatisch dicht; Kelchdruchmesser 5 mm. Gemeine Art in der Oberen Trias der Alpen-Karpatenländer, Pannonien sowie Siebenbürgen (K ő n i g - W a l d - G e b i r g e).

Thecosmilia cf. compressa WEISSERMEL

Fundort: Mecsekgebirge bei P é c s in B á r á n y o s-weger Steinbruch — anisische Schichten. Schlecht erhalten. Kelch typisch meandristisch verlängert *(compressa)* und 26—28 mm. Diastome der Polypenröhren 8 mm. Die Art fand ich in anisischen Schichten der CSSR. Die Art ist schon in Anisikum spezialisiert (meandrinisiert) und kann also schon als eine parakmische festzustellen.

Bavarosmilia bavarica (FRECH)

Fundorten: Budaergebirge bei R e m e t e-Berg und in Zentral-Pannonien bei V e s z p r é m V á r-Berg. Obere Trias. Polypendurchmesser riesig, d. h. 20—30 mm. Polypenrohr kurz (Progression-Regression als phylogenetischer Einheit). Eine üppige Sprossung ist auch festzustellen.

Elysastraea cf. profunda (REUSS)

Taf. II. Fig. 1.

Fundort: L á b a t l a n in Trias-Linse. Kelche tief *(profunda)*, Septen starr und geradlienig. Kelchkonturen rundlich-pentagonal auch hexagonal. Variierend. Eine peripherische Endothekalring ist recht festzustellen.

Palaeastraea decussata (REUSS)
(Syn: *P. incrassata* [SMITH]).

Taf. III. Fig. 2.

Fundort: Budaergebirge bei Farkas-Tal in obertriassischem Kalke (grauem Kalke) eingebettet.

Eine kolonisierte Koralle. Dissepimente vorhanden. Kolumelle fehlt. Epithek und Pseudotheka (letztere zwischen Zentralseptenenden und Epithek) vorhanden. Die Costae sind von Zwischenraum der Epithek und Pseudotheka ausgebildet. So eine richtige Nomenklatur nach KÜHN („Geschiebe von Triaskorallen vom Plabutsch bei Graz" in: Mitt. Nat. Ver. Steierm. 73. 1936. p. 29—32) lautet folgendermassen: „Mauer" richtig: Epithek; „Innenmauer" richtig: Pseudotheka. Von Pseudotheka bis Zentralenden sind die Septen, von Pseudotheka bis Epithek sind die Costae vorhanden.

Coelocaenia borsodi n. sp.

Taf. II. Fig. 6.

Nach Personalnahme Borsod (eine alte hunnische Held-Nahme).

Fundort: Bükkgebirge (Komitat Borsod) in anisischen Schichten. Polypen in grauen Kalke eingebettet und ausgewittert. Gruppenweise vorkommend. Vergleichen wir nun unsere Polypen mit den weissermelschen Angaben zusammen — wie folgt:

Eigenschaften Arten	Polyp	Höhe mm	Diameter mm	Kelch Rand	Septen	Endothekalring	Vorkommen
C. assmanni	Cyl.	60	5—6	glatt	44—48	ist	kolon.
C. exporrecta	Cyl.	?	6—8	?	30—32	ist	kolon.
C. borsodi	Cyl-Trichter	40	9	kamm-artig	90	ist	grupp.

Also die neue Art hat spezifische Charakteristiken wie folgt: *trichteroides* Polyp, grosses Diameter ,kammige Kelchränder, viele Septen. Der Genuscharakter d. h. Endothekalring ist konstant.

Margarosmilia confluens (MÜNSTER)

Taf. I. Fig. 1.

Fundorten: Bükkgebirge bei Zsércnagydél und unter Bánya-Berg, ladinische Stufe. Polyzentrische Gruppen. Das Vorkommen in anisischen Schichten (9) ist zweifelhaft. In Veszprémer Mitteltrias nach PAPP (13) überhaupt sehr dominierend vorkommend. (Syn. *M. zieteni).*

Margarastraea cf. klipsteini FRECH

Taf. I. Fig. 2.

Epidotisch: meandrinisierte Art.

Fundorten: Bükkgebirge: Plateau und Felsőtárkány. In mitteltriassischem Kalke eingebettet und schlecht erhalten. Gemein im Mittertrias von Pannonien (*Veszprém* — PAPP).

Calamophylliopsis (Hymenophyllia) fenestrata (REUSS) KÜHN

Taf. I. Fig. 3, 4, 5.

Fundorten: Budaergebirge bei Remete-Berg; Pestszentlőrinc in eine Triaslinse und Tata, sowie Bükkgebirge.

Diese kolonisierte Korallenart ist auch eine bank- und kalkbildende organogene Faktor der Ursprung der Kalksteine der Oberen Trias. Zwischen Polypenröhren sind wohl entwickelte Kommissuren vorhanden. Kelche mit wohl entwickelten Pali (*Hymenophyllia* — KÜHN 12) die einen Grund geben für Absonderung von anderen koloniebildenden Triaskorallen überhaupt.

Conophyllia recondita recondita (LAUBE)

Fundorten: Bükkgebirge bei Zsércnagydél und Bükk-Plateau. In weissem mitteltriassischen Kalke eingebettet. Gemeine Art. Vorkommen vereinzelt (solo), oder gruppenweise bezw. vergesellschaftet. System der Septen triadisch, d. h. labil, d. h. semiradial und semibilateral mit oft vorkommenden asymmetrischen Septenkonfluenzen. Kolumelle klein. Diameter 2x2 mm (Parakmisch).

Conophyllia recondita pannonica KOLOSVÁRY

Taf. II. Fig. 5.

Fundort: Mecsekgebirge in Misina Gipfel — anisische Recoaro Kalke. Schlecht erhalten. Polypendiameter 7 mm. Akmisch. Septenanzahl 38. Kolumelle klein. Kelchkontur lobulär (desorganisiert.) Siehe Näheres in lit. 9.

Conophyllia omphale KOLOSVÁRY

Fundort: Mecsekgebirge in Misina Gipfel in Recoaro Kalke. Zwei Polypen wurden gefunden eingebettet z. T. ausgewittert. Kolumelle relativ gross, spongiös. Septencyclus 3—4. Septen ungleich entwickelt. Septenanzahl 44. Septen grob und granulär. Kelchdiameter kleiner als 17 mm. Akmisch-riesengewachsen.

„Conophyllia" clepsydrae KOLOSVÁRY

Taf. II. Fig. 4.

Fundort: Mecsekgebirge in M i s i n a Gipfel in Recoaro Kalke. Vorkommen vereinzelt, gruppenweise und kolonie-weise vergesellschaftet. Parakmisch riesengewächsen d. h. Diameter 20—25 mm. Seitensektoren mit starker Reduktion (clepsydriert) und eine Wandvernichtung und Knospung führ zu einer Thamnasterisation über. Durch reiche Knospung und durch das Zusammenbleiben der Polypen führt zu einer Thamnasterisationserscheinung.

Thamnasteria sp. indet.

Fundort: Mecsekgebirge bei P é c s in anisischen Schichten bei dem Französischen Denkmal. Ein sehr schlecht erhaltenes Stückchen.

Thamnasteria cf. alpina WINKLER

Fundort: Bükkgebirge unter B á n y a -Berg.

Eine Art aus der Mitteltrias der Alpenländer, die aber sehr selten zu finden ist. Unser Objekt wegen schlechter Erhaltung schliesst aus einen genaueren Determination.

Anhang

Von mir noch nicht revidierbar, doch als rechte Diagnosen erscheinen noch:

Thamnasteria frechi baconica PAPP (Mitteltrias von V e s z p r é m); *Thamnasteria zitteli* WÖHRMANN (Keszthelyer Gebirge. Obere Trias); *Conophyllia radiciformis* (KLIPSTEIN) (V e s z r p é m, Mitteltrias); *Craspedophyllia cristata* VOLZ, *gracilis* (LAUBE) und *alpina* (LORETZ) (Bakony bezw. Bükkgebirge, Mitteltrias); *Margarosmilia confluens* (MÜNSTER) (V e s z p r é m, dominant im Mitteltrias — PAPP —); *Elysastraea profunda maior* FRECH (Cserhátergebirge. Obere Trias); *Thecosmilia cespitosa* REUSS (Obere Trias); *Montlivaltia septafindens* VOLZ (Keszthelyer-und Bükkgebirge — Obere Trias); *Montlivaltia obliqua* (MÜNSTER) (Keszthelyer Gebirge — Obere Trias).

Zusammenfassung

In Mitteltrias vorherrscht in ungarischen Schichten die Art *Thecosmilia badiotica* (in 7 Fundstellen bankbildend d. h. massenhaft); in der oberen Trias dominiert bei uns die Art *Montlivaltia norica* (ebenfalls von 7 Fundstellen bekannt geworden); nach Artanzahlen ist in U n g a r n auch Mitteltrias am reichsten, da bisher hier 18 Arten vorzufinden geworden waren (in Anisicum 8, in Ladinicum 10). — In anisischen Schichten dominieren die Conophyllien, in Ladinien die

Thecosmilien, Montlivaltien und *Margarosmilia confluens* (PAPP). — In der oberen Trias vorherrschen die Thecosmilien.

Als Endresultat — samt Angaben meiner CSSR und siebenbürgischen Studien — ist es festzustellen, dass nach Artenanzahlen beurteilt: in Ladinicum differenzierte sich die Korallengemeinschaft und in der oberen Trias wurden die kolonisierten Arten progressiv, die Solos dagegen regressiv und die gruppenweise vorkommende Arten weniger regressiv geworden.

Also:

Nach Funsstellen ist die Zusammenfassung wie folgt:

Keszthelyer Gebirge: Montlivaltia obliqua, norica, septafindens und *Thamnasteria zitteli* (Ob. Tr.)

Bakony Gebirge: Montlivaltia norica, Thecosmilia defilippi, clathrata, Bavarosmilia bavarica, Elysastraea profunda, Calamophylliopsis (Hymenophyllia) fenestrata (Ob. Tr.)

Margarosmilia confluens, Margarastraea klipsteini, Conophyllia radiciformis, Thamnasteria frechi baconica, Craspedophyllia gracilis, cristata, alpina (Mitt. Tr).

Lábatlan: Elysastraea profunda, Thecosmilia cespitosa (Ob. Tr).

Tata: Calamophylliopsis (Hymenophyllia) fenestrata (Ob. Tr.).

Budaer Gebirge: Montlivaltia norica, Thecosmilia cespitosa, clathrata, Bavarosmilia bavarica, Calamophylliopsis (Hymenophyllia) fenestrata, Palaeastraea decussata (Ob. Tr.).

Pestszentlőrinc: Calamophylliopsis (Hymenophyllia) fenestrata (Ob. Tr.).

Cserháter Gebirge: Thecosmilia badiotica, clathrata, Elysastraea profunda maior, (Mittl. bezw. Ob. Tr.).

Bükk Gebirge: Plerophyllum cuneatum, Triadophyllum posthumum, Cölocaenia borsodi (Anis.)

Montlivaltia norica, obliqua, legányii, Thecosmilia badiotica, subdichotoma, granulata, Margarosmilia confluens, Margarastraea klipsteini, Conophyllia recondita, Craspedophyllia alpina, Thamnasteria alpina (Mittl. Tr.)

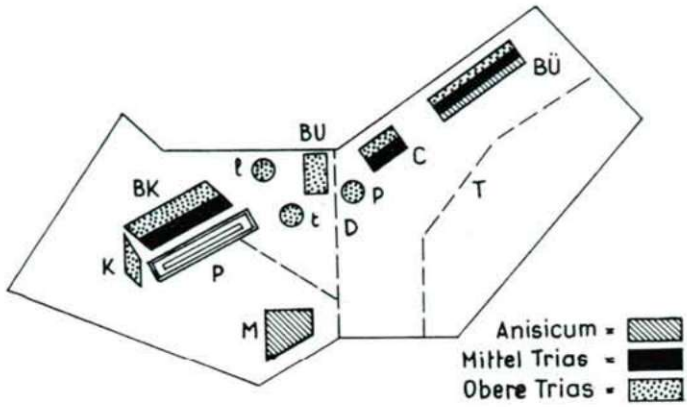
Montlivaltia spetafindens, Calamophylliopsis (Hymenophyllia) fenestrata (Ob. Tr.).

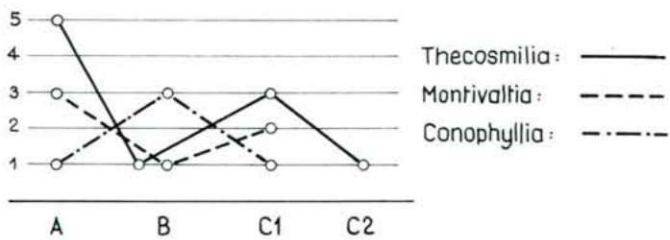
Mecsek Gebirge: Montlivaltia solomoni, Thecosmilia compressa, Conophyllia r. pannonica, omphale, clepsydrae, Thamnasteria sp. (Anis).

Kartenskizze-Erklärung:

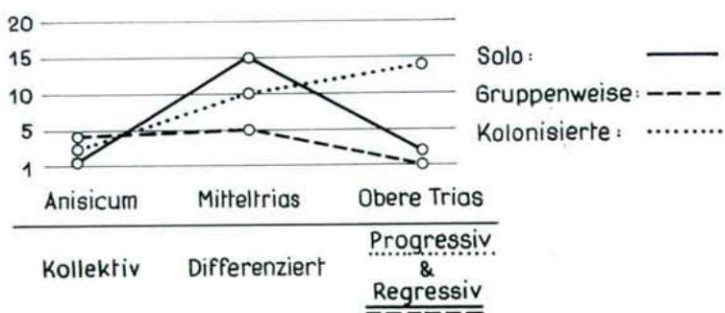
BK Bakony Gebirge	l Lábatlan
M Mecsek Gebirge	p Pestszentlőrinc
BU Budaer Gebirge	t Tata
BÜ Bükk Gebirge	P Plattensee
C Cserháter Gebirge	D Donau
K Keszthelyer Gebirge	T Theiss

Die haupt (dominierenden) Genera *Thecosmilia, Montlivaltia* und *Conophyllia* haben ein vergleichendes Verhältniss in phylogenetischer Hinsicht wie folgt:





Wenn man annimmt, dass die mit konservativen Eigenschaften besitzende Arten von Thecosmililen 5, von Montlivaltien 3, von *Conophyllia* 1, mit elastischen Eigenschaften (grössere Var. und ssp.) 1 *Thecosmilia*, 1 *Montlivaltia* und 3 Conophyllien sind und dass 3 Thecosmilien, 2 Montlivaltien und 1 *Conophyllia* progressiv d. h. besser specialisiert und endlich nur 1 *Thecosmilia* regressiv (*T. defilippi* als desorganisierte Art) erscheint — so sind wir zum folgenden Resultat angekommen:



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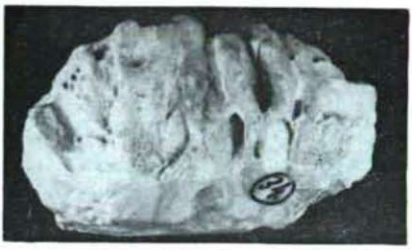

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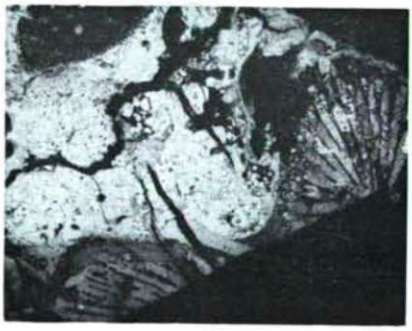

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Taf. I.: 1. *Margarosmilia confluens* Kelche — ausgewittert.
2. *Margarastraea klipsteini* meandrinisierte Oberfläche.
3. *Calamophylliopsis (H.) fenestrata* Kolonie mit Kommissuren — ausgewittert.
4. *Calamophylliopsis (H.) fenstrata* Kolonieteil mit Kommissuren — ausgewittert.
5. *Calamophylliopsis (H.) fenestrata* Kelch mit Pali — gut zu sehen (Schliff).
6. *Thecosmilia cespitosa* Schliff.
7. *Thecosmilia cespitosa* — ausgewittert.

Phot.: Frau PENCZI und L. HAVRANEK


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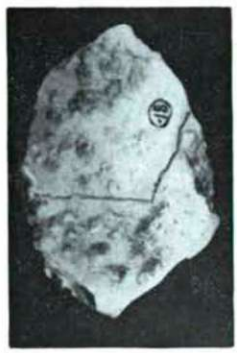
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Taf. II.: 1. *Elysastraea profunda* Schliff.
2. *Plerophyllum cuneatum* Kelchanteil geschlifft.
3. *Montlivaltia cf. solomoni* Seitenansicht eingebettet — ausgwittert.
4. *„Conophyllia" clepsydrae* Kolonie ausgewittert.
5. *Conophyllia r. pannonica* Kelch ausgewittert.
6. *Coelocaenia borsodi* ausgewittert.
7. *Triadophyllum posthumum* Schliff.
8. *Triadophyllum posthumum* Schliff.

Phot.: Frau PENCZI und L. HAVRANEK

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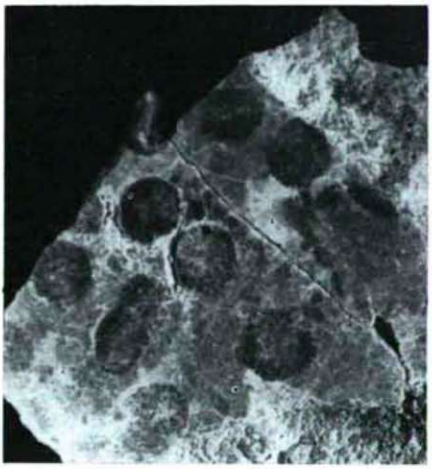

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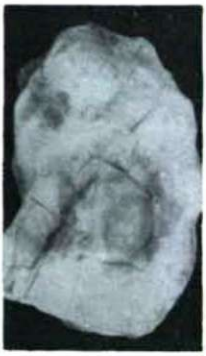

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Taf. III.: 1. *Thecosmilia subdichotoma* Kolonie in situ.
2. *Palaeastraea decussata* Kolonie Schliff.
3. *Thecosmilia granulata* Polypen eingebettet — ausgewittert.
Phot.: L. HAVRANEK

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